



DAA11 HANDBOOK

11th SYMPOSIUM ON DISEASES IN ASIAN AQUACULTURE (DAA11)

23 AUGUST 2022

BCKK, KUCHING SARAWAK

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About this Handbook

Handbook of the 11th Symposium on Diseases in Asian Aquaculture (DAA11) 23rd – 26th August 2022 Kuching, Sarawak

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This volume is the pre-symposium compilation of abstracts. Although oral presentations were selected from submitted abstracts by three independent reviewers, the contents are not peer-reviewed and apart from lay-out changes, the contents have been printed as received from the submitting authors. Some abstracts also need extensive English editing. Please consult with the authors before using information contained in any of the abstracts. The Fish Health Section of the Asian Fisheries Society does not guarantee that this Handbook is without flaws and therefore disclaims any liability for any error, loss or other consequence which may arise from persons relying on the information in this publication. The symposium programme is correct of the time of the printing, however the organizers reserve the right to make changes where necessary.

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1. Selected papers from keynote speaker and participants will be published in Special Issue of Malaysian Fisheries Journal <http://fri.dof.gov.my/>

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Greetings

Dear ladies and gentlemen,

On behalf of the Fish Health Section of the Asian Fisheries Society (FHS-AFS), I am pleased to welcome you to the 11th Symposium on Diseases in Asian Aquaculture (DAA11) Kuching, Sarawak. The early history of the FHS-AFS is closely linked with the Asian Fish Health Network (AFHN), which was initiated in 1985 with the support from the International Development Research Centre (IDRC), Canada. In 1989, the AFHN joined the Asian Fisheries Society (AFS) and the FHS was born as a formal section of the AFS. A year later, the FHS-AFS organized the 1st Symposium on Diseases in Asian Aquaculture (DAA1) in Bali, Indonesia.

What started in Bali in 1990 as a humble gathering has evolved as the biggest fish health event in Asia. Up until DAA10 in Bali in 2017, we managed to hold this symposium every 3 years for 27 years, which is impressive. To celebrate 30 years of the Symposium, we brought DAA11 to Malaysia, where the FHS-AFS was founded in 1989. DAA11 was initially planned to take place in 2020 but postponed to 2022 due to the Covid-19 pandemic. Due to travel restrictions associated with the pandemic, DAA 11 was organized as a virtual event.

DAA11 attracted 629 registered participants and 142 abstract submissions from 22 countries. With the theme of 'Land of Adventure: Exploring Aquatic Animal Health for Sustainable Aquaculture', DAA11 will cover wide ranges of aquatic animal health aspects including biosecurity in aquaculture, epidemiology, detection, prevention, and control of diseases in finfish, crustacean and shellfish. There are also sessions dedicated on trends in fish and shrimp health management. We hope this scientific programme will provide platforms for sharing knowledge, experience, and most importantly lessons learned of managing aquatic animal health in the region and beyond.

DAA11 is jointly organized by the FHS-AFS and the Malaysian Government. On behalf of the FHS-AFS, I would like to sincerely thank the Malaysian Government including the Sarawak Government, Ministry of Agriculture and Agro-Based Industry, and Department of Fisheries for their support and contributions to the organization of DAA11. This event will not be possible without the hard work of the DAA11 Technical Team of the National Organizing Committee. It is my duty to sincerely thank sponsors and exhibitors for their support to the event and to the Executive Committee and Senior Advisors of the FHS-AFS and the International Scientific Committee of DAA11 for their contributions to the development of a strong scientific programme. Last but not least, I would like to extend, my gratitude to the invited speakers and presenters for sharing their work at DAA11. I'm sure your talks will stimulate robust discussion and motivate the participants to further strengthen their networking in Asia and beyond. I hope all of you have a very productive symposium and look forward to seeing you in-person at DAA12 in 2025.

Best wishes,

Dr Agus Sunarto

Chairperson of the FHS-AFS



Welcome Message

Dear Colleague and Friends,

On behalf of the Organizing Committee, I welcome all to the first ever virtual symposium on Diseases in Asian Aquaculture (DAA). While we regret that the COVID pandemic prevented us from holding the symposium in Kuching, we are excited about the opportunities of holding an innovative virtual symposium. The 11th DAA symposium marked the starts of new approaches and strategies, where the current pandemics have reminded us of our interconnectedness, that our health, animal health, and the environment are intrinsically linked - highlighting the urgent need to increase collaboration at the national,

regional, and global levels to address the issue of disease transmission in aquatic ecosystems, zoonotic diseases, antimicrobials resistance as well as aquatic animal welfare, promoting integrated and system based approach to One Health concept.

The DAA11 virtual will includes 4 days of information sharing, intellectual discussions and networking with strategic virtual exhibitions and trade displays covering wide range of topics on aquatic animal health management, development, and innovations presented by internationally recognized experts and researchers from all over Asia Pacific region and around the world. The symposium also provides an excellent platform for international partners to establish and enhance research collaborations, as well as market opportunities for aquatic products and aquaculture technologies. There will be ample time for virtual networking during the Symposium.

The promotion of aquatic animal health management is not just for specialists and scientists but for everyone in the community including farmers with lived experience, aquatic resources managers, veterinarians, aquatic animal health professional, academics, educators, students, all related fisheries industry players, and the general public. Reaching new frontiers requires fresh vision and novel strategies. Producing the symposium online fits perfectly with these aims as it will allow is to keep registration fees to a minimum and reach a wider audience than a traditional conference can possibly include.

I truly appreciate and thank our co-organizing partners, the Fish Health Section of the Asian Fisheries Society (FHS-AFS) and the Ministry of Modernization of Agriculture, Native Land and Regional Development (MANRED); our collaborators especially the Marine Fishery Resources Development and Management Department of the Southeast Asian Fisheries Development Center (SEAFDEC MFRDMD), Universiti Putra Malaysia (UPM), Universiti Malaysia Terengganu (UMT), Universiti Malaysia Sarawak (UNIMAS), International Islamic University Malaysia (IIUM), WorldFish, Malaysia Fisheries Society (MFS) dan Aquaculture Asia Pacific (AAP); and our sponsors; for their technical and financial supports, that has contributed towards the success of DAA11. My sincere appreciation also goes out to all the experts and researchers (who will be participating in the DAA11 Symposium), for their roles in sharing vital and beneficial scientific information as well as valuable experiences on aquatic animal diseases and prevention strategies, contributing and enriching the scientific, public, and private communities. I hope everyone will use these opportunities to enhance and build their capacity towards sustainable, stable, and competitive aquaculture industries for food security and aquatic ecosystem sustainability.

The virtual symposium represents an opportunity for DAA11 to inspire the world with a perspective of hope on research, diagnostics, and services of aquatic animal health in Asian aquaculture. We are certain all of you find this symposium stimulating, rewarding and meaningful.

We are looking forward to warmly welcoming you to DAA11 Virtual

Haji Mohd Sufian Bin Sulaiman

Director General of Fisheries Malaysia

Chairman of the National Organizing Committee for DAA11

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Dr. Liang Yan (P.R. China)
Ms. Varinee Panyawachira (Thailand)

ePoster

Dr. Siow-Foong Chang (Singapore)
Dr. P.K. Pradhan (India)
Dr. Melba G. Bondad-Reantaso (Philippines/Italy)

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Mr. Mohd Faizal Harun (DOF)
Mr. Mazlan Othman (DOF)
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Dr. Rimatulhana Ramly (NaFisH)
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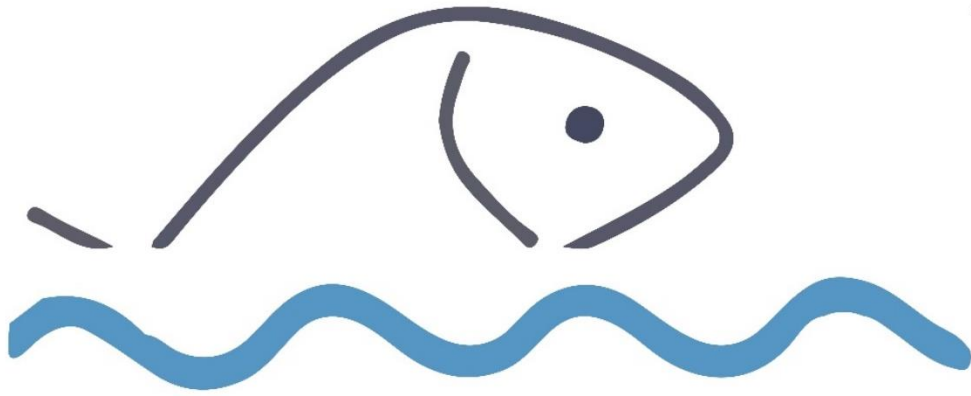
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Program at a Glance

23rd -26th August 2022



DATE	MALAYSIA TIME (MYT), UTC +8	PROGRAM	Mode
DAY 1 23 rd August (Tuesday)	08:00 - 09:45	Registration	Hybrid
	09:50 - 10:30	Opening Ceremony	Hybrid
	10:40 -11:10	Plenary I – Dr. Melba B. Reantaso	Hybrid
	11:30 -17:00	Farmers Day for Malaysian Farmer	Physical
	Session 1: Biosecurity in Aquaculture		
	11:10 - 11:40	Keynote 1 – Dr. Edgar Brun	Virtual
	11:40 - 12:40	Oral Presentation	Virtual
	12:40 - 13:10	3-Minute Presentation	Virtual
	Session 2: Epidemiology (Parasitic, Bacterial and Viral Diseases)		
	14:00 - 14:30	Keynote 2 – Prof. Dr. Kenton L.I. Morgan	Virtual
	14:30 - 15:30	Oral Presentation	Virtual
15:30 - 16:20	3-Minute Presentation	Virtual	
DAY 2 24 th August (Wednesday)	08:30 - 09:00	Plenary II – Dr. Rohana Subasinghe	Virtual
	Session 3: Detection Method/Diagnostic (Parasitic, Bacterial and Viral Diseases)		
	09:00 - 09:30	Keynote 3 – Prof. Karin Pittman	Virtual
	09:30 - 11:30	Oral Presentation	Virtual
	11:30- 12:00	3-Minute Presentation	Virtual
	Session 4: Prevention and Control Measures (Parasitic, Bacterial and Viral Diseases)		
	14:00 - 14:30	Keynote 4 – Dr. Huang Jie	Virtual
	14:30 - 15:45	Oral Presentation	Virtual
	15:45 - 16:00	3-Minute Presentation	Virtual
16:00 – 16:30	Presentation of EoI to Host DAA12 in 2025	Virtual	
Session 5: Trends in Fish Health Management			
DAY 3 25 th August (Thursday)	09:30 - 10:10	Keynote 5 – Dr. Kua Beng Chu	Virtual
	10:10 - 15:00	Oral Presentation	Virtual
	15:30 - 16:00	3-Minute Presentation	Virtual
	16:00 – 19:00	The 12 th Triennial General Meeting of FHS-AFS (TGM12)	Virtual
	Session 6: Trends in Shrimp Health Management		
DAY 4 26 th August (Friday)	09:30 - 10:00	Keynote 6 – Prof. Chu-Fang Lo	Virtual
	10:00 - 12:15	Oral Presentation	Virtual
	14:45 - 15:25	3-Minute Presentation	Virtual
	15:25- 16:00	Presentation of upcoming DAA12	Virtual
	16:00 – 17:00	Awards & Closing Ceremony	Virtual

Scientific Programme Schedule

Plenary	Keynote	Oral Presentation	3-Minute Presentation
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DAY 1: 23 August 2022 (Tuesday)	
SESSION 1: BIOSECURITY IN AQUACULTURE	
08:00 - 09:50	Registration
09:50 – 10:30	Opening Ceremony
Moderator: Dr. Eduardo Leano (Philippines/Thailand)	
10:40 – 11:10	Plenary I: Drivers and pathways to disease emergence in aquaculture and possible solution <i>Melba G. Bondad-Reantaso</i>
Moderator: Dr. Venmathi Maran Balu Alagar (Malaysia)	
11:10 - 11:40	Keynote 1: Biosecurity and sustainability <i>Edgar Brun</i>
11:40 - 11:55	OP-01: Australia's new national aquatic animal disease reporting system <i>Yuko Hood, Joshua Allas, Max de Kantzow and David Gale</i>
11:55 - 12:10	OP-02: Diagnosis in a fish farmer's backpack <i>Jerome Delamare-Deboutteville, Suvra Das, Oleksandra Silayeva, Shaun Wilkinson, Fernando Cagua, Ha Thanh Dong, Saengchan Senapin and Andrew C Barnes</i>
12:10 - 12:25	OP-03: The threat of antimicrobial resistance and solutions in reducing antimicrobials use <i>Ikhsan Natrah, Sarmila Muthukrishnan, Norfarrah Mohamed Alipiah, Md Yasin Ina-Salwany, Chong Chou Min, Low Chen Fei and Nurliyana Muhammad</i>
12:25 - 12:40	OP-04: bioDOF-Map system, a web-based GIS application for mapping fisheries biosecurity activities in malaysia <i>Eleanor Daniella Lokman, Norsida Man and Suhaida Aini</i>
Moderator: Assoc. Prof. Dr. Mohammad Noor Amal Azmai (Malaysia)	
12:40 – 12:55	3MP-01: Occurrence, antibiotic resistance and plasmid profiling of <i>Vibrio</i> spp. isolated from cultured shrimps in Peninsular Malaysia <i>Wan Haifa Haryani Wan Omar, Mohd Zamri-Saad, Mohammad Noor Azmai Amal, Natrah Fatin Mohd Ikhsan and Md Yasin Ina-Salwany</i>

12:40 – 12:55	3MP-02: Fish kill events and harmful algae blooms in peninsular Malaysia waters <i>Roziawati Mohd Razali, Nurin Izzati Mustapa and Harman Muhd-Farouk</i>
	3MP-03: Presence of microplastics in fish parasites: a threat? <i>Najihah Mohamad, Kua Beng Chu, Rohaiza Asmini Yahya, Muhammad Syafiq Musa and Ku Kassim Ku Yaacob</i>
	3MP-04: Antimicrobial resistance surveillance from farmed aquatic foods in wet markets and antimicrobial use prevalence in Bangladesh's aquaculture <i>Shafiq Rheman, Md. Abir Hasan, Laura Khor, Chadag Vishnumurthy Mohan and Jerome Delamare-Deboutteville</i>
	3MP-05: An assessment on the emerging risks associated with aquaculture escapees: the Malaysian case studies <i>Haslawati Baharuddin, Padilah Bakar, Rimatulhana Ramly, Mohamad Sufiyan Salmi, Iftikhar Ahmad Abdul Rafi and Kua Beng Chu</i>
13:00-14:00	Lunch
SESSION 2: EPIDEMIOLOGY	
Moderator: Dr. P. K. Pradhan (India)	
14:00 - 14:30	Keynote 2: Investment, training, resources, rigor and speed a paradigm for aquatic epidemiology <i>Prof. Dr. Kenton L.I. Morgan</i>
14:30 - 14:45	OP-01: In <i>Litopenaeus vannamei</i> peritrophin regulates the pathogenesis of white spot disease and acute hepatopancreatic necrosis disease <i>Ramya Kumar</i>
14:45 - 15:00	OP-02: Description of a Fish Epidemiology and Health Economics (FEHE) survey tool for performance and risk factor assessment <i>Laura Khor</i>
15:00 - 15:15	OP-03: Molecular epidemiology of Megalocytivirus in Malaysian freshwater angelfish (<i>Pterophyllum scalare</i>) <i>Sandra Catherine Zainathan, Che Azarulzaman Che Johan, Muhd Danish Daniel Abdullah and Sharifah Noor Emilia Syed Jamil Fadaak</i>
15:15 - 15:30	OP-04: Characterisation of <i>Edwardsiella ictaluri</i> recovered from clinical disease outbreaks in <i>Pangasianodon hypophthalmus</i> farms in Vietnam over a twenty-year period <i>Christopher Payne, Kathryn Grace, Vo Hong Phuong, Le Hong Phuoc and Margaret Crumlis</i>

Moderator: Dr. Neeraj Sood (India)

15:30 -16:20	<p>3MP-01: Occurences of <i>Vibrio</i> spp. in the samples of cockles (<i>Tegillarca granosa</i>) and Green Mussels (<i>Perna viridis</i>) cultured along West Coast of Johor <i>Azlina Apandi, Fadzilah Yusof, Mohd Lazim Mohd Saif, Azmi Rani and Mohd Farazi Jaafar</i></p>
	<p>3MP-02: TiLV: New threat to Malaysia's native fish species <i>Afzan Muntaziana Mohd Pazai, Fahmi Sudirwan, Shahidan Hashim, Zuraidah Roli, Munira Murni, Suphia Amiera, Mohd Syafiq Mohd Ridzwan and Azila Abdullah</i></p>
	<p>3MP-03: The surveilance of parasitic infestation on green mussels (<i>Perna viridis</i>) in several culture spots in West Coast Johor Water <i>Mohd Lazim Saif, Azmi Rani, Fadzilah Yusof, Azlina Apandi and Mohd Farazi Jaafar</i></p>
	<p>3MP-04: Characterisation of pathogenic <i>Aeromonas</i> species from diseased tilapia and catfish farmed in East Nusa Tenggara, Indonesia <i>Heri Kurniawan, Jimmy F. Turnbull, Andrew P. Desbois and Mags Crumlish</i></p>
	<p>3MP-05: Bacterial microbiota of freshwater prawn, <i>Macrobrachium rosenbergii</i> (de Man, 1879) cultured in cages and ponds in the Philippines <i>Ma-ann Camarin, Erlinda Cruz-Lacierda, Rolando Pakingking Jr., Rex Ferdinand Traifalgar, Nathaniel Añasco and Mark Lawrence</i></p>
	<p>3MP-06: Occurrence of Tilapia Lake Virus (TiLV) in tilapia and tinfoil barbs at wild and culture sites in Malaysia <i>Azila Abdullah, Afzan Muntaziana Mohd Pazai, Mohd Syafiq Mohammad Ridzuan, Munira Murni, Aina Nabila Abdul Rahman, Zuraidah Roli and Rimatulhana Ramly</i></p>
	<p>3MP-07: <i>In-vitro</i> antimicrobial activity of probiotics incorporated with micro feed against pathogenic <i>Vibrio parahaemolyticus</i> <i>Danial Iman, Fadhil Syukri, Siti Hajar Othman and Murni Karim</i></p>
	<p>3MP-08: Infectivity and genome persistence of Tilapia tilapinevirus in water samples <i>Jidapa Yamkasem, Tharinthon Prasartset, Puntanat Tattiyapong, Kwanrawee Sirikanchanad and Win Surachetpong</i></p>
	<p>3MP-09: Global seahorses diseases: A review <i>Marjorie Charam and Kua Beng Chu</i></p>
	<p>3MP-10: Genomes and virulence factors of novel <i>Pseudomonas koreensis</i> strain Isolated from aquaculture farm in Kuching, Sarawak, Malaysian Borneo <i>Cindy Jia Yung Kho, Melinda Mei Lin Lau and Hung Hui Chung</i></p>

DAY 2: 24 August 2022 (Wednesday)	
SESSION 3: DETECTION METHOD/DIAGNOSTIC (PARASITIC, BACTERIAL & VIRAL DISEASES)	
08:00 - 08:30	Registration
Moderator: Dr. Agus Sunarto (Indonesia/Australia)	
08:30 - 09:00	Plenary II: State of global aquaculture <i>Rohana Subasinghe</i>
Moderator: Dr. Yuko Hood (Australia)	
09:00 - 09:30	Keynote 3: Fish wear their immune system on the outside-what this means for aquaculture and ecology <i>Pittman Karin, Merkin Grigory, Okubamichael Mearge, Powell Mark, Andersen Linda and Carlo C. Lazado</i>
09:30 - 09:45	OP-01: Profile of protein isolated from mucus of infected <i>Oreochromis</i> spp <i>M. Faizal, M. Nur-Nazifah, M.R. Syafiq, G. Wan Norazlan, A. Azila and R. Rimatulhana</i>
09:45 - 10:00	OP-02: Rapid CRISPR-based platforms for detection of major pathogens in penaeid shrimp and fish <i>Thawatchai Chaijarasphong, Suthasinee Kanitchinda, Thanwarat Sukonta, Thanyawit Thammachai, Saengchan Senapin, Kallaya Sritunyalucksana, Pimkhuan Hannantanan, Jiraporn Srisala, Rungkarn Suebsing, Watcharachai Meemetta, Suwimon Taengphu and Mintra Kitthamarat</i>
10:00 - 10:30	Virtual Tea break
10:30 - 10:45	OP-03: Application of a specific antibody to localize Tilapia Lake Virus (TiLV) in fish tissues using immunohistochemistry <i>Puntanat Tattiyapong, Chutchai Piewbang, Somporn Techangamsuwan and Win Surachetpong</i>
10:45 - 11:00	OP-04: An intelligent protozoan white spot fish disease detection <i>Siti Naquiah Md Pauzi, Amiera Syazlin Md Azhar, Nor Hazlyna Binti Harun, Mohamad Ghozali Hassan, Noraini Yusoff and Kua Beng Chu</i>
11:00 - 11:15	OP-05: Gut metagenome analyses of healthy and diseased tilapia <i>Oreochromis niloticus</i> cultured in Bangladesh <i>Mohammad Shamsur Rahman, Nusrat Jahan Punom, Mst. Sabrina Akter, Md. Mostavi Enan Eshik, Shawon Ahmmed, Md Golam Rabbane and Mst. Khadiza Begum</i>
11:15 - 11:30	OP-06: Glucose and glutamine contribute to de novo nucleotide synthesis to support viral pathogenesis <i>Cong-Yan Chen, Der-Yen Lee, Shih-Shun Lin, Chun-Hung Liu and Han-Ching Wang</i>

Moderator: Dr. Ng Wing Keong (Malaysia)	
11:30 – 12:00	<p>3MP-01: Mass mortalities of golden pomfret (<i>Trichinotus blochii</i>) associated with oxygen crisis, multiple infections of parasites and <i>Vibriosis</i> at floating cages Pulau Aman, Penang <u>Padilah Bakar, Rimatulhana Ramly, Fahmi Sudirwan and Kua Beng Chu</u></p>
	<p>3MP-02: Experimental co-infection of Tilapia Lake Virus and <i>Streptococcus agalactiae</i> in red hybrid tilapia (<i>Oreochromis niloticus</i> × <i>O. mossambicus</i>) model <u>Lukman Basri, Mohammad Noor Amal Azmai, Mohd Zamri Saad, Ina Salwany Md. Yassin, Annas Salleh and Yasmin Abd Rahman</u></p>
	<p>3MP-03: Investigation of culture condition inducing viable but non-culturable (VBNC) state in <i>Nocardia seriolae</i> <u>Akito Hayashi, Megumi Matsumoto, Motohiko Sano and Goshi Kato</u></p>
	<p>3MP-04: Mucosal responses in giant freshwater prawn, <i>Macrobrachium rosenbergii</i> fed with diet containing natural feed premix <u>Imelda Rantty, Siti Rokhaiya Biollah, Karin Pittman, Kho Li Yung and Siti Hawa Mohamad Ali</u></p>
	<p>3MP-05: Experimental challenges studies on pathogenicity of <i>Edwardsiella ictaluri</i> in striped catfish, <i>Pangasianodon hypophthalmus</i> (Sauvage) <u>Amira Syahidah Nordin, Mubarak Muhyuddin Noor Zainy, Muhammad Ansarullah Mohd Subari and Nur Nazifah Mansor</u></p>
	<p>3MP-06: Pathogenicity and LD₅₀ of <i>Pangasius nasutus</i> against <i>Edwardsiella ictaluri</i> through injection method <u>Syafiq Syauqi and Nur Nazifah Mansor</u></p>
	<p>3MP-07: <i>In-vivo</i> expression of Acute Hepatopancreatic Necrosis Disease related virulence genes in <i>Vibrio parahaemolyticus</i> and <i>V. harveyi</i> and its virulence towards <i>Penaeus vannamei</i> (Boone, 1931) <u>Sarmila Muthukrishnan, Mohamed Shariff, Ina-Salwany M. Y, Fatimah Md Yusoff and I. Natrah</u></p>
	<p>3MP-08: Phenotypic characterization of <i>Edwardsiella ictaluri</i> Isolated from cage-cultured <i>Pangasianodon hypophthalmus</i> <u>Rimatulhana Ramly, Nur Erina Syahira Abd Talib, Amira Syahidah Nordin and Nur Nazifah Mansor</u></p>
	<p>3MP-09: PvRaf knockdown using RNA interference increased survival of <i>Litopenaeus vannamei</i> infected with White Spot Syndrome Virus <u>Joseph Carlo V. Vergej and Mary Beth B. Maningas</u></p>
	<p>3MP-10: <i>In-vitro</i> evaluation of potential probiotics strain in biofilm formation <u>Sow Cyn Shieng, Fatimah Md Yusoff, Chong Chou Min, Jasmin M.Y and Murni Karim</u></p>
12:00 - 14:00	Lunch

DAY 2: 24 August 2022 (Wednesday)	
SESSION 4: PREVENTION & CONTROL MEASURES (PARASITIC, BACTERIAL & VIRAL DISEASES)	
Moderator: Dr. Sandra Catherine Zainathan (Malaysia)	
14:00 - 14:30	Keynote 4: Emerging aquatic animal diseases and response actions to their emergencies in Asia and the Pacific Region <i>Jie Huang</i>
14:30 - 14:45	OP-01: Towards heuristic design of vaccines: Transforming transposon insertion sequencing (TIS) for identification of novel <i>in-vivo</i> virulence factors in fish pathogen <i>Qiyao Wang</i>
14:45 - 15:00	OP-02: Application of natural products: an effective approach to control fish parasites in aquaculture <i>B. A. Venmathi Maran</i>
15:00 - 15:15	OP-03: In silico selection and characterization of DNA aptamers against <i>Vibrio parahaemolyticus</i> LuxP periplasmic receptor protein <i>Low Chen Fei, Siti Aisyah Razali, Nur Afiqah Md Yusof, Azyyati Mohd. Padzil, Syarul Nataqain Baharum, Nurul Hanun Ahmad Raston, Chong Chou Min, Natrah Fatin Mohd Ikhsan, Sarmila A/P Muthu Krishnan and Magdalena Lenny Situmorang</i>
15:15 - 15:30	OP-04: Potential practical application of a live attenuated vaccine against herpesviral hematopoietic necrosis of goldfish <i>Hiroaki Saito, Shungo Minami, Manami Yuguchi, Aiko Shitara, Hidehiro Kondo, Goshi Kato and Motohiko Sano</i>
15:30 - 15:45	OP-05: Active and passive immune responses and protection of Nile tilapia (<i>Oreochromis niloticus</i>) immunized with Tilapia Lake Virus inactivated vaccines <i>Thao Thu Mai, Pattanapon Kayansamruaj, Suwimon Taengphu, Chayanit Soontara, Pattarawit Kerddee, Dinh-Hung Nguyen, Saengchan Senapin, Janina Z. Costa, Jorge del-Pozo, Kim D. Thompson, Channarong Rodkhum and Ha Thanh Dongi</i>
Moderator: Dr. Chang Siow Foong (Singapore)	
15:45 – 16:30	3MP-01: Organic acid profiles of three potential probiotics from whiteleg shrimp, <i>Litopenaeus vannamei</i> <i>Jikon Alvion J. Denwinner, Ang Chun Yao, Annita Yong Seok Kian, Lim Leong Seng, Sujjat Al-Azad, Francis Yong and Mohammad Tamrin Mohamad Lal</i>
	3MP-02: Field assessments of oral vibrio vaccine indicate the potential for protection against Vibriosis in hybrid grouper, <i>Epinephelus fuscoguttatus</i> × <i>Epinephelus lanceolatus</i> <i>Aslah Mohamad, Md Shirajum Monir, Mohd Zamri Saad, Mohammad Noor Amal Azmai, Nurhidayu Al-saarie and Ina-Salwany Md Yasin</i>
	3MP-03: Field efficacy of a feed-based inactivated <i>Vibrio harveyi</i> vaccine against vibriosis in cage-cultured asian seabass, <i>Lates calcarifer</i>

15:45 – 16:30	<i>Zahaludin Amir Danial, Aslah Mohamad, Mohd Zamri Saad, Mohammad Noor Amal Azmai, Annas Salleh and Ina-Salwany Md Yasin</i>
	3MP-04: <i>In-vitro</i> antimicrobial activity of plant extracts against fish pathogenic bacteria <i>Siti Hawa Mohamad Ali, Imelda R., Kho LY., Siti Rokhaiy B. and David Y.</i>
	3MP-05: Control of <i>Aeromonas</i> sp infection in nursery stage of <i>Pangasius nasutus</i> (Bleeker, 1863) through Bio-encapsulation of Herbal Extract; SirehMax in Live Feed <i>Iftikhar Ahmad A. R., Hanan M. Y., M. Zudaidy J. and Nik Haiha N. Y</i>
	3MP-06: Assessment of inhibitory properties of potential probiotics isolated from <i>Aeromonas hydrophilla</i> and <i>Streptococcus agalactiae</i> <i>Shafiq Johar</i>
	3MP-07: Ultrafiltration membrane technology for virus mitigation control strategy in indoor shrimp farming system <i>Norhafiza Ilyana Yatim, Diyana Kamarudin, N. Ali, S.C. Zainathan and S.N.K. Addis</i>

DAY 3: 25 August 2022 (Thursday)	
SESSION 5: TRENDS IN FISH HEALTH MANAGEMENT	
09:00 - 09:30	Registration
Moderator: Dr. Supranee Chinabut (Thailand)	
09:30 - 10:00	Keynote 5: Malaysian experiences: Environment-friendly alternatives to chemicals in aquatic animal health management <i>Kua Beng Chu, Padilah Bakar, Ahmad Baihaqi Othman, Rohaiza Asmini Yahya and Wan Norhana Md. Noordin</i>
10:00 - 10:15	OP-01: IgY as immunoprophylaxis in aquaculture <i>Chou Min Chong, Phek Har Chua, Mohd Shafiq Mohd Nor, Nur Syahirah Yahya and Natrah Fatin Mohd Ikhsan</i>
10:15 - 10:30	OP-02: The effects of oral feed-based vaccination against vibriosis on gut microbiome of Asian seabass <i>Jumria Sutra, Amalia Mohd Hashim, Mohd Termizi Yusof, Nurhidayu Al Saari, Ina Salwany Md Yusof and Mohammad Noor Amal Azmai</i>
10:30 – 11:00	Virtual Tea break
11:00 - 11:15	OP-03: Ectoparasites recovered from whole cage freshwater treatment on cultured marine fish disease outbreak in floating cages <i>Leong Tak Seng</i>

11:15 - 11:30	OP-04: The prospect of using aptamers and digital aptasensors for real-time surveillance of diseases in aquaculture <i>Tang Kok Mun</i>
11:30 - 11:45	OP-05: Zoonotic potential of Group B Streptococcus in aquaculture <i>Snehashish Snehashish, Ina Salwany Md Yasin, Syafinaz Amin Nordin and Mohammad Noor Amal Azmai</i>
11:45 - 12:00	OP-06: Phytobiotic-based additive for consistently improved gut integrity and health across species <i>Ruth Garcia Gomez, M. M. Isern Subich and W. G. Nuez Ortinc</i>
12:00 – 12:15	OP-07: Refolded recombinant major capsid protein (MCP) from Infectious Spleen and Kidney Necrosis Virus (ISKNV) effectively stimulates serum specific antibody and immune related genes response in Nile tilapia (<i>Oreochromis niloticus</i>) <i>Sarocho Jitrakorn, Boonyalit Throngnumchai, Triwit Rattanarojpong and Vanvimon Saksmerprom</i>
12:15 - 14:00	Lunch
14:00 - 14:15	OP-08: Effects of <i>Piper betle</i> extract supplementation diet as a natural antibiotic growth promoter (NAGP) to kelah (<i>Tor tambroides</i>) <i>Hanan Mohd Yusof, Ahmad Baihaqi Othman, Nik Haiha Nik Yusoff, Muhd Zudaidy Jaapar, Azila Abdullah, M Firdaus Nawawi and Nur Nazifah Mansor</i>
14:15 - 14:30	OP-09: In-vitro responses of peritoneal macrophages of marine red hybrid tilapia (<i>Oreochromis spp.</i>) model to vibriosis: A comparative study between vaccinated and non-vaccinated fish <i>Tilusha Manchanayake, Ina Salwany Md. Yassin, Mohammad Noor Amal Azmai, M. Zamri Saad, Mohd Firdaus Nawawi and Annas Salleh</i>
Moderator: Dr. Andy Shinn (United Kingdom/Thailand)	
14:30 – 15:15	3MP-01: The Effect of Different Concentration of Probiotics (Lacto-sacc) Mixtures on Growth Performance and Feed Utilization of Empurau (<i>Tor tambroides</i>) Fingerlings <i>Sing Ying Chua, Roslianah Asdari and Mohammad Bodrul Munir</i>
	3MP-02: The effect of commercial cinnamon essential oil (EOCIN) against bacterial infection in farmed red snapper (<i>Lutjanus argentimaculatus</i>) <i>Noor Hanis Abu Halim, Padilah Bakar, Rohaiza Asmini Yahya, Nurhidayati Ahmad Sobri, Wan Rozana Wan Ahmad and Kua Beng Chu</i>
	3MP-03: The Management of Reared Tiger Grouper (<i>Epinephel us fuscoguttatus</i>) Infected with Marine Leech (<i>Zeylanicobdella arugamensis</i>) <i>Fadzilah Yusof, Azmi Rani, Mohd Lazim Saif Azlina Apandi and Mohd Farazi Jaafar</i>
	3MP-04: Preliminary study of water quality in relation to benthic organism under aquaculture cages situated in Rambungan River, Lundu <i>Kho Li Yung, Imelda R, Siti Hawa MA, David Y and Siti Rokhaiya B</i>
	3MP-05: Dietary lacto-sacc improved growth performance and basic haematological parameters in empurau (<i>Tor tambroides</i>) fries reared in the aquaponics system

	<i>Mohammad Bodrul Munir, Sharifah Lia Farliana Wan Alias and Roslianah Asdari</i>
14:30 – 15:15	3MP-06: Field evaluation of palm oil adjuvanted feed-based streptococcosis vaccine efficacy in red hybrid tilapia <i>Mohd Syafiq Mohammad Ridzuan, Azila Abdullah, Mohd Firdaus Nawawi, Norazsida Ramli and Hanan Mohd Yusof</i>
	3MP-07: Disinfection of rotifer, <i>Brachionus plicatilis</i> leading to bacteria-free populations using SirehMAX™ solution <i>Nur Fatin Afifah Osman Manah, Mohd Salleh Hasan, Nur Atikah Azmi, Shaharah Mohd Idris, Azila Abdullah and Nur Nazifah Mansor</i>
	3MP-08: Development of acquired immune system in striped catfish <i>Pangasianodon hypophthalmus</i> <i>Dung Nguyen Ngoc, Oanh Dang Thi Hoang, Motohiko Sano and Goshi Kato</i>

DAY 4: 26 August 2022 (Friday)	
SESSION 6: TRENDS IN SHRIMP HEALTH MANAGEMENT	
09:00 - 09:30	Registration
Moderator: Prof. Dr. Kallaya Sritunyalucksana (Thailand)	
09:30 - 10:00	Keynote 6: The quorum sensing (QS) system modulates virulence of <i>Vibrio parahaemolyticus</i>^{AHPND} by regulating the expression of the <i>pirA^{VP}</i>/<i>pirB^{VP}</i> genes through AphB^{VP} <i>Chu-Fang Lo, Shin-Jen Lin and Hao-Ching Wang</i>
10:00 - 10:15	OP-01: <i>Piper betle</i> L. leaf extracts inhibit quorum sensing of shrimp pathogen <i>Vibrio harveyi</i> and protect <i>Penaeus vannamei</i> postlarvae against bacterial infection <i>John Paul Matthew D. Guzman, Pattanan Yatip, Chumporn Soowannayan and Mary Beth B. Maningas</i>
10:15 - 10:30	OP-02: Involvement of white spot syndrome virus viral proteins in modulating shrimp glycolysis <i>Yen Siong Ng, Saengchan Senapin, Pakkukul Sangsuriya and Han-Ching Wang</i>
10:30 - 10:45	OP-03: Shrimp GDH may be regulated by SIRT4 during WSSV replication <i>Tan Yu Kent and Han-Ching Wang</i>
10:45 – 11:15	Virtual Tea break
11:15 - 11:30	OP-04: WSSV-induced metabolic reprogramming in shrimp hemocytes to benefit its re0lication <i>Han-Ching Wang</i>

11:30 - 11:45	OP-05: Quorum quenching activity in microalgae and its associated bacteria: an alternative antimicrobial therapeutics <i>Sarmila Muthukrishnan, Natrah Fatin Mohd Ikhsan and Nur Ain Yahya</i>
11:45 - 12:00	OP-06: Microbiome signatures from stocking to emergency harvest in Thailand <i>Penaeus vannamei</i> pond experienced Yellow Head Virus outbreak <i>Anuphap Prachumwat, Wiraya Eamsaard, Piyanuch Wechprasit, Apirak Hoonlor, Diana Minardi, Grant D. Stentiford and Kallaya Sritunyalucksana</i>
12:00 - 12:15	OP-07: Dietary montmorillonite clay protected <i>Penaeus vannamei</i> challenged with <i>Vibrio parahaemolyticus</i> (AHPND) and modulated stomach microbiota of shrimp Survivors <i>Wing-Keong Ng, Mei-Ling Mong and Abdul Azim Abdul Hamid</i>
12:15 - 14:45	Lunch
Moderator: Dr. Chadag Vishnumurthy Mohan (India/Malaysia)	
14:45 – 15:25	3MP-01: The effectiveness of probiotic Used in super intensive culture of white shrimp (<i>Penaeus vannamei</i>) for its health and water quality management <i>Azmi Rani, Fadzilah Yusof, Mohd Lazim Saif Azlina Apandi and Mohd Farazi Jaafar</i>
	3MP-02: Preliminary study on the effect of moist formulated feed on health status of disease infected <i>Penaeus monodon</i> broodstock <i>Mohammed Suhaimie Abd Manaf, Mohd Firdaus Azmi, Nor Aida Suzana Abdul Rahman and Rosnani Yaakub</i>
	3MP-03: Blue-green algae and assessment of microcystin in shrimp aquaculture farms of Sarawak <i>Lim Mui Hua</i>
	3MP-04: Scientometrics of climate change and shrimp disease: An overview <i>Mohamad N. Azra, Mohd Ihwan Zakariah, Mohd Iqbal Mohd Noor and Mazlan Abd Ghaffar</i>
	3MP-05: Microsporidian parasite, <i>Enterocytozoon hepatopenaei</i> at early stage of culture of whiteleg shrimps (<i>Peneaus vannamei</i>) in West Malaysia <i>Rohaiza Asmini Yahya, Padilah Bakar, Wan Muhammad Hazim Wan Sajiri, Nur Samihah Mazlan, Nur Ashikin Arbi and Kua Beng Chu</i>
	3MP-06: Unveiling the potential of medicinal plants in combating shrimp Acute Hepatopancreatic Necrosis Disease (AHPND) disease <i>Tan Wen Chian, Nisha Krishnan, Loh Jiun Yan, Baskaran Gunasekaran and Hok Chai Yam</i>
15:25 - 16:00	Presentation of upcoming DAA12



DAY 1: 23 August 2022 (Tuesday)
Session 1: Biosecurity in Aquaculture

Plenary I

Drivers and pathways to disease emergence in aquaculture and possible solution

Melba G. Bondad-Reantaso

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Globally, the trend in aquaculture is that a serious transboundary infectious disease of aquatic organism emerges, spreads rapidly, and causes major production losses approximately every three to five years. The many factors contributing to disease emergence in aquaculture can be categorized into four major pathways, including: (i) trade in live animals and products; (ii) knowledge of pathogens and hosts; (iii) health management and disease control; and (iv) ecosystem changes. Emerging diseases in aquaculture may occur due to the introduction of new diseases through various exposure pathways, or due to an increased impact of diseases already present. The consequences of an emerging disease are dependent on the time of detection and effectiveness of response to the disease, which rely on good capacities in surveillance, diagnostics, institutional coordination, research infrastructure, biosecurity, and disease control measures. National strategic planning on aquatic health management and biosecurity is vital to reduce the vulnerability of the aquatic sector to new and emerging diseases and the often *ad-hoc* and reactive solutions to disease outbreaks and mass mortality events in aquatic populations.

The Progressive Management Pathway for Improving Aquaculture Biosecurity (PMP/AB) developed by FAO and partners, is a 'paradigm shift' that follows the principles of being risk-based, progressive and collaborative, and focuses on diseases faced by aquaculture at the commodity and enterprise levels. The four stages of PMP-AB involve strong stakeholder input to promote the application of risk management as part of a national approach. Countries decide the appropriate entry-point, how far and how fast to progress to the next stage. Due to the wide variation of farmed aquatic species, aquaculture sectors may advance independently, at different speeds or with different goals but a common requisite is strong cooperation between government, industry and academic sectors, under the public-private sector partnership (PPP). This is necessary to ensure clarity on roles and responsibilities, identify key gaps requiring improved capacity and infrastructure, and increase awareness of the cost/benefits of biosecurity systems. Emergency preparedness and risk analysis are key aspects of all stages of the PMP/AB. Risk hotspots (critical control points) along the value chain are identified for biosecurity investment (training, diagnostic capacity, etc.). All these feed into development of national strategy on aquatic organism health or aquaculture biosecurity, which sets the foundation for ongoing review and updating as the industry develops.

As countries and aquaculture enterprises advance along the pathway, the PMP/AB is expected to: reduce the burden of diseases, improve aquatic (organism and environmental) health at the farm and national levels, minimize international spread of diseases, improve socio-economic benefits from aquaculture, increase investment in aquaculture, and achieve One Health goals; all of which provide benefits at the enterprise, national, regional, and global levels.

Short Biography

Dr Melba B. Reantaso, Ph.D. (Ms), has close to 30 years combined experience in research, training, diagnostics, extension and international aid and development work on aquaculture, management of health of aquatic organisms and biosecurity. She retired early as Senior Aquaculturist in 2002 after 18 years of government service at the Fish Health Section of the Philippine Bureau of Fisheries and Aquatic Resources. Dr Reantaso worked for the Network of Aquaculture Centre in Asia and the Pacific (NACA, Bangkok, Thailand) from 1999-2002 as Regional Aquatic Animal Health (AAH) Specialist and Coordinator of an FAO project with 21 countries participating that resulted to the establishment of aquatic animal health programme of NACA; and led the investigation of koi herpesvirus, at that time, in Indonesia. She migrated to the USA and worked (2002-2004) at the Cooperative Oxford Laboratory, a fish health laboratory shared by MD/DNR and NOAA, responsible for the histopathological analyses from an active surveillance for oysters diseases. She joined FAO in 2004, led international disease investigation task forces on epizootic ulcerative syndrome in southern Africa and the Democratic Republic of Congo, white spot syndrome virus of shrimp in Mozambique and Madagascar and acute hepatopancreatic necrosis disease in Viet Nam. She has M.Sc., Ph.D. and post-doc credentials all on diseases of aquatic animals: post-doc fellow (Nippon Veterinary and Life Science University, adviser: Prof Kishio Hatai) of the Japan Society for the Promotion of Science; Ph.D. (University of Tokyo; adviser: Prof Kazuo Ogawa) as a Monbusho scholar and an M.Sc in Biology (De La Salle University, adviser: Dr James Richard Arthur) as a recipient of a project scholarship grant from International Development Research Center of Canada. She earned her B.Sc. in Zoology from the University of the Philippines in Los Banos. She serves as referee to various peer-reviewed journals and co-editor of Diseases in Asian Aquaculture series (DAA V, VI and VII). She was founding member and served as Chairperson (2002-2005) and Secretary/Treasurer (1999-2002) of the Fish Health Section, Asian Fisheries Society and currently Senior Adviser. She travels extensively (at least 68 countries to-date) in pursuit of scholarly and career goals in aquaculture development and biosecurity. She maintains a global network of experts, and served as Chief Editor of the FAO Aquaculture Newsletter from 2006 to 2016. She spearheaded the development of a new initiative called Progressive Management Pathway for Improving Aquaculture Biosecurity, a paradigm shift in managing health of aquatic species through a risk-based, collaborative and progressive approach. She is Team Leader of Food Safety, Nutrition and Health team at the Fisheries and Aquaculture Division of FAO since 2021; aside from supervisory tasks, she leads FAO work on biosecurity, disease risk assessment, surveillance, national/regional aquatic organisms health strategies, AMR in aquaculture; develops and implements capacity development activities, projects, training courses, seminars/conferences; conducts investigations of disease outbreaks and mass mortality events in aquatic populations; and represents FAO in relevant bodies, committees, advisory board, etc..

Keynote I

Biosecurity and sustainability

Edgar Brun

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During the last decades, the global aquaculture industry has grown rapidly, and today provides more aquatic animal products for consumption than traditional fisheries. The development in aquaculture is associated with new technology, high number of species for farming, global demand for food and live animals and genetic products; a trade that connects farmers all over the world. However, the downside of this “success” is a global spread of a variety of infectious diseases with devastating impacts on single farmers, industry, local communities and indeed the environment and the national economics. Knowledge of and a proper understanding of possible impacts of infectious diseases in aquatic animals have been lacking.

This understanding is slowly emerging by an increasing international attention from OIE and FAO as both organizations have adopted biosecurity as a strategic approach to reduce risk and sustain the industry. Biosecurity including prevention of the spreading of infectious agents and an adequate control of disease occurrence is increasingly seen as essential components of any talk about sustainable aquaculture production. Coastlines or inland rivers and lakes connect farms whether they are owned by a multinational company or a small local farmer. A possible interaction between farms therefore exists and the infection status and implementation of biosecurity measures on a farm, a region and a nation, is consequently not a private issue. Rather it is a concern at national and community level, as well as for the industry as such. This puts responsibilities on both the industry as well as the relevant authorities to collaborate, support, guide, report, and regulate the industry. The talk will highlight some of the essential components to succeed in this collaboration aiming to develop a more sustainable industry with respect to infectious diseases.

Keywords: Biosecurity, sustainability, aquaculture

Short Biography

Dr. Edgar Brun is currently working as Director for Aquatic Animal Health and Welfare at the Norwegian Veterinary Institute. He has a Doctor Science degree from the Norwegian School of Veterinary Science, and a master in epidemiology from London School of hygiene and Tropical Medicine/Royal Veterinary College. He worked as a private fish health veterinarian in the growing salmonid industry in Norway before he started his early research carrier in epidemiology at the Norwegian Veterinary Institute where he later became leader for the epidemiology section for 12 years. His interests are various aspects of aquatic animal epidemiology and health management.

Oral Presentation

Australia's new national aquatic animal disease reporting system

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Australia is obligated to report about Australia's status of OIE listed diseases to the OIE World Animal Health Information System. Australia has also agreed to report about its status of regionally reportable diseases to the Asia-Pacific region through the Network of Aquaculture Centre of Asia-Pacific and the OIE Regional Representation for Asia and the Pacific. To fulfil the international obligation and for effective national coordination of information, Australia developed the Quarterly Aquatic Animal Disease system (QAAD). QAAD is an online based reporting system that was developed to collate information from all jurisdictions regarding cases of reportable aquatic animal diseases. It has since become outdated and no longer supported by our contemporary information security system. AUSPestCheck™ is a data sharing and coordination software system, initially developed and managed by a joint government-industry non-profit organisation, Plant Health Australia (PHA), for the purposes of plant health surveillance. PHA, in conjunction with the Department of Agriculture, Water and the Environment (DAWE), initiated trials to use AUSPestCheck™ as a terrestrial animal health surveillance data management system. Recently, the aquatic team of the DAWE, in collaboration with PHA, created "National Aquatic Animal Health Surveillance Trial" in AUSPestCheck™. The suitability of this trial program for collating national aquatic animal disease information for international reporting purposes is being assessed. Through the trial implementation, benefits of this new system have been demonstrated. These include faster, improved uploading and data management process, more streamlined and real-time reporting, improved visualised outputs, and improved access to national disease intelligence that supports decision making. The presentation will provide an overview of Australia's aquatic animal disease reporting system, governance and functions of aquatic AUSPestCheck™. Some case studies will be provided as to how aquatic AUSPestCheck™ would facilitate the sharing, collation and presentation of aquatic animal disease surveillance data for policy decision makers.

Keywords: AUSPestCheck™, OIE, disease reporting, surveillance, surveillance data

Short Biography

Dr Yuko Hood is a Principal Science Officer in the Aquatic Pest Health Policy team within the Australian Government Department of Agriculture, Water and the Environment. Yuko holds a Bachelor of Veterinary Science degree obtained in Japan. Yuko takes a lead role in developing Australia's national aquatic animal health management and diagnostic policies, designing national surveillance projects, and coordinating national emergency aquatic animal disease responses such as white spot disease. Yuko is Australia's OIE focal point for aquatic animals, actively engaging with various international and regional stakeholders on aquatic animal health issues.

Diagnosis in a fish farmer's backpack

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Fish underpin future nutritional security, supplying high quality protein, iron, iodine and vitamin A that are critical to childhood development and deficient in many staple foods. In 2018, 54.1 million tonnes of fish were produced by farming, generating US\$138.5 billion and directly employing 19.3 million people, mostly in developing nations. With expansion and intensification, disease losses are increasing and are a priority for the FAO sub-committee on aquaculture. In most developing countries, disease mitigation comprises over-stocking to compensate, and use of readily available antibiotics. Indeed 67 different antimicrobials are used in the 11 major producing countries, contributing to the global pool of antimicrobial resistance (AMR). Accurate identification of the causes and sources of infectious disease is essential for implementation of evidence-based treatment, biosecurity and prevention. Pathogen genomics can provide sufficiently detailed information but has, to date, been too expensive and time consuming. Lab-in-a-backpack uses nanopore sequencing technology and low-cost, low-waste sample preparation to generate whole pathogen genome sequence data from diagnostic samples on the farm without laboratory support. Our simplified safe workflow includes a cloud-based identification tool that returns near real-time information about the pathogen using any laptop or smartphone. This enables evidence-based treatment, epidemiological tracing, AMR surveillance and the production of simple low-cost locally produced 'autogenous' vaccines to protect the next crop. These big-data-informed but locally implemented solutions align well with FAO's recently proposed Progressive Management Pathway for Improving Aquaculture Biosecurity, and can deliver real advances in local economy, nutritional security, antimicrobial stewardship and animal welfare.

Keywords: Oxford Nanopore Technologies, bacterial genomic, cloud-based identification tools, serotyping, multi-locus sequence typing

Short Biography

Jérôme is a Scientist based in Penang and employed by WorldFish. In this position, Jérôme is working on aquatic animal health issues focusing on tilapia, carp and catfish. His research activities focus on epidemiology, antimicrobial resistance, and the development of novel rapid genomic detection methods of aquaculture pathogens. Before joining WorldFish in June 2018, Jérôme lived in Australia for 10 years where he worked on multiple vaccine research projects for warm water fish species at the University of Queensland (UQ). In 2014 at UQ, he received his PhD on an emerging bacterial disease affecting wild fish species in Australia.

The threat of antimicrobial resistance and solutions in reducing antimicrobials use

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Intensive farming increases stress on the host and introduces new diseases or pathogens to emerge. This scenario has increased the usage of antibiotics in aquaculture worldwide. Antimicrobial drugs can be very useful in combating pathogenic bacterial infections in humans and animals. However, it has negative implications as it increases the number of anti-microbial resistance (AMR) micro-organisms that can spread from animal to human by direct exposure or consumption of food containing AMR. In aquaculture systems, antibiotics are generally added in the feed or directly into the water system. Once, the antibiotics get into the water; it starts to disperse evenly by the aid of the paddlewheel aerators and impose a selective pressure, which eventually changes the ecosystem of the environment. Consequently, some bacteria increase their fitness in the new environment via intrinsic resistance or acquired resistance. In this paper, the threat and potential risk of AMR particularly in shrimp aquaculture will be discussed. This includes potential mitigation strategies to reduce AMR involving quorum quenching mechanisms and others.

Keywords: Antibiotic, diseases, aquatic animal, and communication

Short Biography

Dr. Natrah Ikhsan is an Associate Professor in the Department of Aquaculture, Faculty of Agriculture, Universiti Putra Malaysia (UPM). She is also an Associate Researcher in the Aquatic Animal Health and Therapeutics Laboratory, Institute of Bioscience, UPM and an Interim Member of Laboratory of Sustainable Aquaculture, International Institute of Aquaculture and Aquatic Sciences, UPM. Currently, she is the Deputy Dean of Graduate studies, Research and International, Faculty of Agriculture, UPM. She was the Treasurer of World Aquaculture Society-Asian Pacific Chapter and Vice President, Malaysian Fisheries Society. Dr. Natrah specializes in the field of Aquatic Microbial Ecology particularly in the development of innovative and sustainable microbial management strategies through understanding of the host-microbe interaction for enhanced microbial stability. Among her current research focus is on quorum sensing sociomicrobiology – its pathogenic and probiotic role particularly in bacteria, algae & other hosts in aquaculture ecosystem.

bioDOF-Map system, a web-based GIS application for mapping fisheries biosecurity activities in Malaysia

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A web-based system provides access to a software system using a computer, tablet, smartphone with an internet connection and based on a type of spatially distributed information. Their relationship is penetrated over a network, expressed through URLs (created by the server) and HTTP. This study is to design, develop, produce, emplace, test and deliver, together with proper attestation, user manual and training for web-based GIS application with User Acceptance Testing (UAT) in increasing the use of remote sensing technology and other technologies such as Geographic Information System on the management and monitoring of fisheries biosecurity activities. The system developed based on the DOF-Malaysian Space Agency (MYSA) Memorandum of Understanding (MoU), to simplify mapping and navigation, can be used with the Internet such as Google Chrome. There are five modules, first module is view for display list of basic information and aquaculture farms information, second module was search widget and third Module is edit widget. Fourth Module is a measurement widget. The last module is the printing widgets. The system can be explored by using smartphones and tablet that supports Android, iOS, and Windows phone. This system can also know the distribution of the disease status based on surveillance results which are being updated by the fisheries officers online and the notifications will be highlighted in dashboard. The system module can be expanded with an analysis module in the future. The system empowers clients to utilize the GIS data set with farm information and other services, without needing a piece of profound information in the field of GIS or utilizing GIS programming. This system helps the management in planning, developing facilities and services to the stakeholders in Malaysia. It also to reducing the use of human resources in the management, giving important data on fisheries, aquaculture and fish disease in Malaysia.

Keywords: GIS, Remote Sensing, web-based mapping, spatial

Short Biography

Ms. Eleanor Daniella binti Lokman is currently working as Senior Research Officer at the Department of Fisheries Malaysia. She is currently pursuing her Ph.D in Agricultural Extension in University Putra Malaysia. She received her Master of Science in Rural Development from the University of Putra Malaysia. She then worked at the Malaysian Remote Sensing Agency, Ministry of Science, Technology and Innovation (MOSTI), and served as a Research Officer at the Agriculture Application Unit. She has authored several publications in various journals and books. Her publications reflect his research interests in GIS, Remote Sensing, Fisheries, Social Science and Agricultural Extension”.

3-Minutes-Presentation

Occurrence, antibiotic resistance and plasmid profiling of *Vibrio* spp. isolated from cultured shrimps in Peninsular Malaysia

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Shrimps are major seafood in the aquaculture sector, particularly in the Southeast Asia. However, *Vibrio*-related infections frequently occur in shrimp farms. Antibiotics are widely used in shrimp farming to prevent or treat disease outbreaks. Unfortunately, the use of antibiotics may lead to antibiotic resistance among animals and humans population. Moreover, the information on the occurrence of *Vibrio* spp. and antibiotics use in shrimps, particularly in Malaysia, is minimal. Thus, the study aims to provide information on the occurrence and distribution of the antibiotic resistance and plasmid profiling patterns isolated from cultured shrimps in Peninsular Malaysia. Shrimps were sampled from seven farms located in different geographical regions in Peninsular Malaysia. Then, the isolates were biochemically tested and subjected to molecular detection using the *pyrH* gene. The isolates also were screened for their antimicrobial resistance against 16 antibiotics and plasmid profiling. A total of 225 isolates belonging to 13 different *Vibrio* species were successfully isolated and characterized, which are *V. parahaemolyticus* (55%), *V. communis* (9%), *V. campbellii* (8%), *V. owensii* (7%), *V. rotiferianus* (5%), *Vibrio* spp. (4%), *V. alginolyticus* (3%), *V. brasiliensis* (2%), *V. natriegens* (2%), *V. xuii* (1%), *V. harveyi* (1%), *V. hepatarius* (0.4%), and *P. damsela* (3%). Antibiotic resistance profiles showed that most isolates were resistant to penicillin G (100%) but susceptible to norfloxacin (96%). The MAR index ranged from 0.06 to 0.75. Analysis of isolates revealed that 16% with MAR index less than 0.2 while 84% of the isolates with MAR greater than 0.2. 56% of the *Vibrio* isolates harbored plasmid with molecular weight ranging from 1.0 to 10kb were detected among the resistant isolates. After the curing process, the multiresistant isolates lost their antibiotic resistance phenotype to several antibiotics. The most frequent was cephalothin (KF). At the same time, few isolates remained resistant to tetracycline (TET) and sulfamethoxazole-trimethoprim (SXT). This suggested that the isolates were mediated by both chromosomal and plasmid mediation among the resistant isolates. The finding will support surveillance data and information on the occurrence and emerging trends of antimicrobial-resistant and plasmid profiles in shrimp farms.

Keywords: Shrimps, *Vibrio* spp, antibiotic resistance, plasmid, Peninsular Malaysia

Short Biography:

Wan Haifa Haryani is a Ph.D. student at Universiti Putra Malaysia. She completed her MSc (Biochemistry) from Universiti Kebangsaan Malaysia in 2012. Currently, she is a special graduate research assistant at UPM, working on microbial biotechnology and aquatic biotechnology, focusing on shrimps. Wan-Haifa Haryani has authored several publications in various journals and proceedings.

Fish kill events and harmful algae blooms in Peninsular Malaysia waters

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Harmful algal blooms (HABs) are a natural phenomenon that has the potential to cause mass mortality on fish from factors such as lower oxygen levels in waters (anoxia/hypoxia) or excretion of toxic bioactive compounds that may clog, irritate or damage the fish gills and lead to suffocation. HAB is well-known to be associated with mass mortality of cage-cultured fish and had caused recurrent events in northern Peninsular Malaysia and the straits of Johor waters. These HABs have caused huge losses for the aquaculture industry. This status review gives a summary of HABs events in Peninsular Malaysia associated with a fish kill from the year 2000 to end of the year 2020. There were some identified phytoplankton species that could cause fish mortalities from its bloom in northern Peninsular Malaysia including *Ceratium furca* (in Perak on March 2007 and June 2008), *Margalefidinium polykroikodes* (in Perak on February 2013 and 2014), *Noctiluca scintillans* (in Perak on March 2016 and June 2020) and *Margalefidinium fulvescens* (in Perak and Penang on May 2020), causing massive losses for the fish farmer. Meanwhile, a bloom of *Karlodinium australe* have threatened caged fish in West Johor Straits in February 2014 and recurred in February 2015. Thus, regular phytoplankton monitoring need be implemented to provide early warning and minimize the impact of HAB incidents in Peninsular Malaysia especially in areas where HAB events are frequently reported. In addition, there should be an awareness program to fish farmers about HABs and their negative impact on the economy and health to safeguard the mariculture industries.

Keywords: cage culture fish, fish mortality, fish killer microalgae, Peninsular Malaysia

Short Biography:

Roziawati Mohd Razali is currently working as Research Officer at Fisheries Research Institute. She received her Master degree on Marine Science from the UNIMAS, Sarawak. Roziawati Mohd Razali has authored several publications in various journals. Her publications reflect his research interests in marine microalgae especially harmful microalgae.

Presence of microplastics in fish parasites: A threat?

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Microplastics are pervasive pollutants in the marine environment and poses threat to food chain and yet no study has reported any occurrence of microplastics in ectoparasites of marine fishes. The purpose of this study was to investigate the occurrence of microplastics in ectoparasites found from cultivated fish in Pulau Pinang, Malaysia. Ectoparasites were wholly digested as a group with each (n=10) according to species and microplastics were further identified using a stereomicroscope. A total of 33 microplastics were found in this study with an average of 1.1 particles/individual and the smallest size recorded was 11.02 μ m. This finding manifested that plastic particle are bioavailable in the fish tissue which may provide a huge risk for fish and towards humans consuming fish. Additional comprehension of microplastics exposure towards fish and its associated additives is needed to determine the appropriate permissible level of microplastics in fish for fish health and human consumption.

Keywords: Microplastics, Ectoparasites, Marine fishes, Occurrence, Bioavailable

Short Biography

Najihah is as a research officer at the Fisheries Research Institute (FRI) Batu Maung Pulau Pinang, Malaysia. She received her Bachelor's degree on Applied Science (Fisheries) from the University Malaysia Terengganu. Najihah is currently pursuing her Master of Ocean Science from Universiti Kebangsaan Malaysia. She then worked at Malaysian Agricultural Research and Development Institute (MARDI) served as a research officer at the Centre for Marker Discovery and Validation (CMDV). Najihah has authored several publications in various journals and newsletters. Her publications reflect his research interests in marine pollution specifically microplastics.

Antimicrobial resistance surveillance from farmed aquatic foods in wet markets and antimicrobial use prevalence in Bangladesh's aquaculture

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Antimicrobial resistance is a globally recognized One Health problem imposing major threats on public health management of infectious diseases. Aquaculture is one of the leading food sector that is challenged by infectious diseases leading to irrational and inappropriate use of antimicrobials. Antimicrobial resistant pathogens from aquaculture can find their way to terrestrial animals, human and their environments, and vice versa, thereby affecting the whole ecosystem. To generate the evidence of antimicrobial resistance in key pathogens from farmed aquatic foods, we sampled 240 fish/shrimp specimens from four large wet markets in Dhaka and Khulna Bangladesh. We successfully isolated and identified four bacterial groups of human significance (*E. coli*, *Salmonella* spp. *Vibrio* spp. and *Streptococci* spp.) from different organs of the sampled specimens. We performed antimicrobial susceptibility testing for a panel of 16 antibiotics. Our findings confirmed the presence of antimicrobial resistant bacteria in fish and shrimp at point of human consumption. We also performed whole genome sequencing of selected bacterial isolates and derived predicted antimicrobial resistant genes and virulence factors. To characterize antimicrobial use prevalence and identify related factors in commercial aquaculture, we used a structured questionnaire and interviewed 192 aquaculture farmers, 4 livestock-input dealers and 20 purchasing customers covering two southern districts of Bangladesh, Khulan and Satkhira. Our observations coincide with previous findings of wider usage of antimicrobials in Bangladesh aquaculture, with variation between farming systems and regions. We argue that factors associated with antimicrobial use in Bangladesh's aquaculture are also embedded in socioeconomic heterogeneity.

Keywords: One Health, Aquaculture, antimicrobial resistance, antimicrobial use, Antimicrobial susceptible testing

Short Biography

Mr. Shafiq Rheman is currently working as Scientist at the WorldFish. He received his Master's degree from the University of Hull, UK. He worked for a number of reputed organizations including Food and Agriculture Organization, Bangladesh as Fisheries Consultant. Mr. Rheman has authored several publications in various journals. His publications reflect his research interests in antimicrobial resistance and One Health.

An assessment on the emerging risks associated with aquaculture escapees: the Malaysian case studies

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Aquaculture is the world's fastest growing food-producing sector and currently the main source of fish supply. Global aquaculture relies on the farming of non-native aquatic species. These features of aquaculture, combined with large-scale movements of animals, have driven disease emergence, with major threat to biodiversity and ecosystem function. Escapees from aquaculture facilities can result in aquatic bioinvasions and there is increasing evidence of their impacts on human health and economies globally. In this review, case studies associated with aquaculture-based fish mortalities and diseases, reported to NaFisH and Department of Fisheries Malaysia (DoF) from 2007-2020, are analysed. Disease investigation was initiated from reports or complaints of fish mortalities and disease problems occurring at aquaculture farms by farmers, private sectors, government institutions or fish mass mortalities in waterbodies by general public. Annual case reported varies from as low as 1-3 cases per year (2008, 2014 and 2016) to the highest of 62 cases in 2010. A total of 216 cases were reported with Tilapia (52.3%), African Catfish (21.3%), River Catfish (11.1%), Whiteleg Shrimp (5.5%) and Koi (5.1%) are the main species infected. Closed culture systems (tanks and ponds) contributed to 60.2% compared to open culture systems (cages in rivers and lakes – 39.8%), with earthen ponds contributed to the highest number of cases (33.3%). About 54.6% cases were diagnosed with bacterial infections caused mainly by *Aeromonas* spp. (26.4%), *Vibrio* spp. (11.6%) and *Staphylococcus* spp. (10.7%) while infection by parasites contribute to 12% and virus (17%). Tilapia Lake Virus (TiLV) was the only viral disease reported, mostly infect Tilapia. This analysis highlights the urgent need for better preventive and mitigation measures to reduce fish escapes, particularly in sensitive biodiversity areas, considering risk assessment for farming non-native species and the critical role of policy makers in implementing these measures to allow the sustainable aquaculture development.

Keywords: Aquaculture, non-native species, bioinvasion, biodiversity, biosecurity

Short Biography

Dr. Haslawati is currently working as a senior researcher at the Freshwater Division, Fisheries Research Institute (FRI) Glami Lemi, Jelebu, Negeri Sembilan. She received her Doctoral degree or PhD on Fisheries Sciences from the University of Malaya, Malaysia. She then worked at the Department of Fisheries, Malaysia, for nearly 25 years. Dr. Haslawati has authored several publications in various journals and books. His publications reflect his research interests in sustainable aquaculture and biodiversity conservation, especially related to invasive alien species.

DAY 1: 23 August 2022 (Tuesday)
Session 2: Epidemiology



Keynote 2

Investment, training, resources, rigor and speed a paradigm for aquatic epidemiology

Kenton Morgan

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“Epidemiology, pandemic, R number;” words and concepts that were once the trademark of epidemiologists became part of everyday language as a result of global infection with an RNA virus. The last 2-3 years has taught us those longstanding methods of infectious disease control i.e. the identification of infected individuals or populations and their isolation from non-infected individuals or populations remain the basis of infectious disease control. It has been tough, and it remains tough at personal, national and international levels. It stands as a testimony to the power of infectious diseases and the role of epidemiology in their control. In aquaculture, two major pandemics have occurred over the last 50 years. White Spot Disease Syndrome and Epizootic Ulcerative Syndrome or Mycotic Granulomatosis. There was neither the epidemiological knowledge, infrastructure, finances nor political will to prevent the spread of these diseases. The use of the now obsolete word epizootic is testimony to this. Arguably too much investment was made in trying to identify the “causes” of both and too little investment in their control. Investment is key to infectious disease control. You cannot simply switch on a manufacturing line to produce the expertise and infrastructure needed to combat a pandemic. The people, structures, equipment and importantly networks need to be in place. These networks are about people, who understand each other and get along with one another. Training in epidemiology often focuses on the theoretical and it can be daunting to even the most experienced laboratory scientist involved in infectious diseases. Like all sciences it has its jargon and definitions. It is important that these are understood and used correctly. Confounder for example is a word thrown around in conversation very often with little reference to its actual meaning. Epidemiology however is a practical subject, whether observational, experimental, or theoretical, it is firmly rooted in practical disease control. There is a tendency for it to become top heavy, with theoretical epidemiologists outnumbering “welly boot” epidemiologists. Both are vital for effective disease control. Training should be practical and useful; it should not be a showcase for the complexity of epidemiology and the prowess of epidemiologists. Epidemiologists are often viewed as “bucket scientists” by those based in the laboratory. They use large numbers. These large numbers extend to the resources needed but unlike laboratory science it is often human resource rather than reagents that are needed. People - trained people - on the ground, collecting data, controlling movement, testing vaccines. These are key individuals. Epidemiology is about disease in populations, but it recognises that even single individual and consignments of animals can introduce disease into a previously non- infected waterbody or island country. Where rivers cross national boundaries, disease control should based on catchments not countries and the political structures which enable this should be in place. It is not rocket science! It just involves key people and groups talking to each in a task driven, tolerant way. Where procedures, such as risk-based certification or more draconian exclusion measures are in place, they have to be rigorously observed and maintained. When that “weekend” moment of disease introduction, outbreak or spread occurs, speed is of the essence. Better to put in movement controls and then relax them because of false

alarms rather than inform everyone that borders will be closed on Monday. The effects of hours at this early stage can be massive as are the political and economic pressures on the decision makers! Whilst the control of infectious disease can involve generic and specific preventive measures, the control of non-infectious diseases presents greater challenges to the epidemiologist. One often missed part of the armoury in the control of non-infectious diseases is precedent and analogy. Animal kept in relatively controlled environments and fed manufactured diets grow faster. The demands of more rapid growth on nutrient and micronutrient requirements have resulted historically in diseases of deficiency and imbalance in pigs and poultry. The knowledge that these have occurred in other intensified species should serve as a resource and early warning system to the aquatic epidemiologist. Throughout the world, the provision of epidemiological expertise in aquaculture is woefully inadequate. Purse holders and political decision makers, in countries where aquaculture makes a significant contribution to human health and or GDP fail to invest, often large sums of money, in Epidemiology at their peril!

Keywords: Epidemiology, infection, diseases, control, policy, intensification, aquaculture

Short Biography

Kenton Morgan was the first Professor of Epidemiology at a British Veterinary School. He is a veterinarian with a postgraduate degree in mucosal immunity. A graduate of the University of Cambridge, he developed his interest and skills in epidemiology and practical preventive medicine at the University of Bristol. He was introduced to Aquaculture after a Scotland v Wales rugby game, by Professor James Turnbull of the University of Stirling. Together with Drs CV Mohan and Nguyen Van Hao, they pioneered the application of quantitative epidemiology to aquaculture, in India and Vietnam. He has held epidemiology courses in the USA, Europe, India, and Japan, where he was visiting professor at Nagasaki University. His novel and hands on teaching techniques have attracted international acclaim.

Oral Presentation

In *Litopenaeus vannamei* peritrophin regulates the pathogenesis of white spot disease and acute hepatopancreatic necrosis disease

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Recurrent outbreaks of white spot disease (WSD) and acute hepatopancreatic necrosis disease (AHPND) remain a threat to the shrimp industry worldwide. The shrimp stomach is the primary site for the ingested pathogens, namely white spot syndrome virus (WSSV) and the AHPND causing *V. parahaemolyticus*. However, molecular mechanisms involved in the pathogen attachment and colonization in shrimp stomach remains unclear. Here we investigated the role of *Litopenaeus vannamei* peritrophin gene (*LvPT*) in regulating the pathogenesis of WSD and AHPND. Peritrophic membrane (PM) lines the shrimp digestive tract and peritrophins are PM proteins that can interact with chitin fibres via chitin-binding domains. After WSD or AHPND infection, there was upregulation of *LvPT* mRNA expression. The dsRNA-mediated gene silencing of *LvPT* followed by WSSV challenge significantly reduced expression of *IE1*, *VP28*, *ICP11* genes and WSSV genome replication. In addition, during AHPND infection, *LvPT* gene silencing increased expression of AHPND-associated plasmid and toxin. Overall, in WSSV infection, *LvPT* silencing favoured the host, whereas during AHPND, *LvPT* may regulate bacterial colonization. Although WSSV virions strongly bound to the recombinant *LvPT*, their precise role in disease pathogenesis still needs to be elucidated. These findings provided further insights into host-pathogen interactions and could be useful for developing potential strategies to minimize disease outbreaks.

Keywords: Peritrophin, peritrophic membrane, AHPND, WSSV, *L. vannamei*

Short Biography

Dr. Ramya Kumar is currently working as Post-doctoral Researcher in the Department of Biotechnology and Bioindustry Sciences, National Cheng Kung University, Taiwan. She received her PhD on the role of bile acids in regulating the virulence of AHPND-causing *V. parahaemolyticus* in white shrimp, from the National Cheng Kung University, Taiwan. Dr Kumar completed her MS in Biotechnology from the Bangalore University, India. She has authored several publications in high-ranking SCI journals, with many focusing on identifying key factors in shrimp that mediate host-pathogen interactions in AHPND and WSSV pathogenesis.

Description of a Fish Epidemiology and Health Economics (FEHE) survey tool for performance and risk factor assessment

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Disease control efforts by farmers and national governments are normally hindered by lack of data on the status of fish health, risk factors, and the economics of losses due to disease. To fulfil the need for an integrated assessment of fish epidemiology and health economics in Asia and Africa, the Fish Epidemiology and Health Economics (FEHE) survey tool was created with the objective of collecting baseline data on existing farm practices, production, inputs, epidemiology, losses and economic impacts due to disease and mortalities. The FEHE tool involves the use of the Open Data Kit (ODK) Collect mobile application to access the survey questionnaire in remote farm locations with limited internet connectivity. The questionnaire is stored on KoboToolbox, a free, open source online platform for managing surveys and collecting field data. Using the survey tool, epidemiological findings were obtained from 550 farms in Bangladesh and 110 farms in Egypt. Survey responses were analyzed to identify risk factors and to understand the performance of local aquatic food systems including economic impact of infectious diseases. Findings will be further used to develop targeted risk mitigation interventions and national aquatic animal health strategies for sustainable aquaculture in the targeted regions. A contextualized version of the tool has been recently used in Nigeria as part of the United States Agency for International Development Feed the Future Innovation Lab for Fish (USAID FIL) project and will also be applied in Ghana and Kenya under the Norad supported projects. For the purpose of capacity building, a mobile friendly online course ('Aquaculture survey with ODK') has been created on Learn.ink platform to train survey tool users. The tool and its accompanying guidelines have been described by WorldFish in an Aquatic Animal Health Package of Practices (POPs): Fish Epidemiology and Health Economics.

Keywords: Farm survey tool, Aquaculture baseline survey, Aquaculture risk factor, Fish epidemiology study, Aquaculture performance

Short Biography

Laura Khor is currently working as a Research Fellow at WorldFish Center, Penang. She received her Masters in Aquaculture from Universiti Malaysia Sabah. She then served as a Quality Control Manager at a private aquaculture company in Malaysia for 11 years before joining her current organization. Laura has research interests in aquatic animal health.

Molecular epidemiology of *Megalocytivirus* in Malaysian freshwater angelfish (*Pterophyllum scalare*)

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Malaysia has more than 630 culturists that are involved in the ornamental fish industry for culturing 250 species including local and exotic species. However, megalocytiviruses have been associated globally with severe systemic disease and economic loss in ornamental fish. The intensity of *Megalocytivirus* infection in *Pterophyllum scalare* is unknown in Malaysia. Thus, the objectives of this study include the detection of the presence of *Megalocytivirus*, determination of genotypes and the major risk factors that contribute to precipitation of disease outbreaks in *P. scalare* and lastly, the evaluation of non-invasive sampling detection (swabbing method) in *P. scalare* broodstock. A total of six pairs of freshwater angelfish broodstock were used. The water samples such as inlet river, inlet well, broodstock ponds, breeding pails, growth out ponds, outlet water, and other samples such as mucus swabs, gill swabs, freshwater angelfish eggs, fries, juvenile, snail, snail eggs, live feed (tubifex worms and moina), sediment samples (from river, broodstock ponds, growth out ponds, outlet) and wild fish were collected from the farm from day 0 until day 60. These samples were extracted and proceeded using nested polymerase chain reaction (PCR) analysis according to Whittington et al. (2009). A total of 159 samples included both inlet river and well, broodstock ponds, breeding pails, growth out ponds, outlet water sample, mucus and gill swabs, the eggs fries and juvenile of *P. scalare*, snail and snail eggs, tubifex worm, moina and all of sediment samples showed positive results for *Megalocytivirus*. In conclusion, the results showed the existence of different possible routes of *Megalocytivirus* distribution in the farm.

Keywords: detection, *Megalocytivirus*, ornamental fish, PCR, epidemiology

Short Biography

Assoc. Prof. Dr. Sandra Catherine Zainathan is currently working as lecturer and researcher at the Faculty of Fisheries and Food Sciences, Universiti Malaysia Terengganu, Malaysia. She is also the Programme Head for Bachelor in Aquaculture Science in her faculty. She received his Doctoral degree or PhD on Aquatic Virology from the University of Tasmania, Australia. Assoc. Prof. Dr. Sandra Catherine Zainathan has authored several publications in various journals and books. His publications reflect his research interests in viral diseases in aquatic organisms including finfish, oysters, mussels, shrimps and coral ecosystem.

Characterisation of *Edwardsiella ictaluri* recovered from clinical disease outbreaks in *Pangasianodon hypophthalmus* farms in Vietnam over a twenty-year period

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Edwardsiella ictaluri, the causative agent of bacillary necrosis of *Pangasius* (BNP), has been a significant challenge to the farmed catfish industry in Vietnam since it was first reported over two decades ago. However, little is known about the current heterogeneity of *E. ictaluri* strains circulating Vietnamese striped catfish (*Pangasianodon hypophthalmus*) farms and how these populations have changed over time. Therefore, the aim of this study was to characterize *E. ictaluri* strains associated with BNP in farmed striped catfish in Vietnam over a twenty-year period. A total of 233 strains were investigated in this study, which were recovered from clinical disease outbreaks across six provinces in Vietnam between 2001 and 2021. Bacterial strains were characterized using primary identification methods and their sensitivity to six antibiotic compounds used in global aquaculture profiled. A subset ($n = 83$) of bacterial strains were further typed using various molecular methods. In general, *E. ictaluri* strains were found to be homogenous in their biochemical profiles, irrespective of geographical location or year of recovery. Antibiotic resistance was found to increase over time, with high-level, multidrug resistance associated with strains recovered between 2018 and 2021. Pulse-field gel electrophoresis differentiated selected strains into 16 pulsotypes. Further, strains recovered between 2001 and 2011 were found to cluster distinctly from those recovered after 2017. Screening of virulence genes using PCR methods revealed six conserved virulence mechanisms in selected strains, however differences were noted in the presence of *eseJ* and *traD*, associated with the type IV and IV secretion systems, respectively. Continued surveillance of *E. ictaluri* in Vietnam will be important to inform biosecurity strategies and aid in the development of effective vaccines against BNP in striped catfish.

Keywords: Striped Catfish, *Edwardsiella ictaluri*, Vietnam, Virulence, Antibiotic Resistance

Short Biography

Dr. Payne is currently working as post-doctoral research fellow at the University of Stirling. He received his Doctoral degree or PhD in Aquaculture and Master of Science in Aquatic Pathobiology from the University of Stirling. He has worked as a research assistant on several projects with various fish species including catfish, salmonids, and tilapia. Dr. Payne has authored several publications in various journals, which reflect his research interests in aquatic bacterial pathogens, particularly antimicrobial resistance and the dynamic relationship between host, microbe and the aquatic environment.

3-Minutes Presentation

Occurrences of *Vibrio* Spp. in the Samples of Cockles (*Tegillarca Granosa*) and Green Mussels (*Perna Viridis*) Cultured along West Coast of Johor

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Fresh fish and shellfish still could generate peril for the consumer because they are popularly pick out to be eaten raw or undercooked. As filter feeder, shellfish normally can have threatened the consumers with foodborne illness. Other than harmful alga, bacterial infestation can also develop susceptible food poisoning. During the last few years, there have been some reports on foodborne outbreaks and cases attributed to pathogenic *Vibrio spp* in shellfish. The genus *Vibrio* contains at least twelve species that pathogenic to human, ten of which can cause foodborne illness. The majority of foodborne illness are caused by *V. parahaemolyticus*, *Vibrio cholerae*, or *Vibrio vulnificus*. From the general guidance on the interpretation of results for specific foodborne pathogens in ready-to-eat food stated that the occurrence of *V. cholerae* in 25grams of samples or *V. parahaemolyticus* in $>10^3$ are potentially injurious to health and/or unfit for human consumption. This study aimed to detect the presence of *Vibrio spp* in samples of cockles and green mussels cultured along west coast of Johor. Samples of blood cockles and green mussels were obtained from five areas along west coast of Johor. Total plate count was determined from total amount of the colony-forming-unit (cfu) of the bacteria. Colonies of *Vibrio spp* were isolated from blood cockles and green mussels' samples using *Vibrio* TCBS (Triosulphate bile salt agar) selective media. The result of total plate count bacteria was < 20 colony/g. The entire 40 samples of blood cockles and 50 samples of green mussels were positive containing *Vibrio* which means they were possibly harmful for human consumption if serves raw or undercooked.

Keywords: foodborne; blood cockle; green mussels; *Vibrio*; bacteria

Short Biography

Ms Azlina binti Apandi is currently working as Assistant Research Officer at the Fisheries Research Institute Gelang Patah, Johor, Malaysia. She completed his Diploma of Science in Microbiology from the University Technology MARA. She has 10 years experience in aquaculture and biosecurity.

TiLV: New threat to Malaysia's native fish species

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The global tilapia industry is threatened by an emerging virus called Tilapia Lake Virus (TiLV), also known as syncytial hepatitis of tilapia that was first discovered in Kinneret Lake, Israel. Usually, tilapia is the main host of TiLV but other fish species also shows susceptibility. There is a worry that this virus can infect other species and cause outbreaks. Our immediate concern is its impact to our native species since research found that tilapia is the most dominant alien species in our freshwater ecosystem. An epidemiology study was conducted at Timah Tasoh Lake, Perlis that TiLV was previously reported to check for the presence of TiLV in black tilapia (*Oreochromis niloticus*) and tinfoil barb (*Barbonymus schwanenfeldii*) from March 2018 until March 2020. A total number of 291 *O.niloticus* and 475 *B.schwanenfeldii* were sampled. Organ such as brain, liver, kidney and spleen were pooled in viral transport media (VTM) for cell culture and tested using semi-nested RT-PCR. TiLV prevalence was calculated using online EpiTools software from AusVet. Results showed that the prevalence of TiLV in both species were consistently detected throughout the study period. The result reveals that the prevalence of TiLV in *B.schwanenfeldii* was tested higher (46.99%) compared to *O.niloticus* (32.16%) even in asymptomatic fish. This study shows that TiLV found in these two species is associated with virus circulating in the environment. The epidemiology study suggests that TiLV has already broken the species boundary and potentially could infect other fish species than *B.schwanenfeldii*. This emerging disease could be a threat to our native species since changes in the environmental parameters could trigger a massive fish kill. This will lead to devastating effects on our native species especially waters that are negatively impacted with invasive alien species. Targeted surveillance for TiLV in other fish species should be considered in order to prepare for possible future scenario where TiLV can infect new host species.

Keywords: Tilapia Lake Virus (TiLV), *Oreochromis niloticus*, *Barbonymus schwanenfeldii*, alien species

Short Biography

Afzan Muntaziana Mohd Pazai is currently working as Research Officer at the Fisheries Research Institute Glami Lemi, Negeri Sembilan, Malaysia. She completed her Master Degree of Science in Aquaculture from the Universiti Putra Malaysia. She then worked at the Universiti Teknologi Malaysia (UTM), served as Research Officer at the UTM Innovation Centre in Agritechology for Advance Bioprocessing (ICA). She has authored several publications in various journals and books. Her publications reflect her research interests in Aquaculture.

The surveillance of parasitic infestation on green mussels (*Perna Viridis*) in several culture spots in West Coast Johor Water

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Green mussel, *Perna viridis* is an economically important bivalves and food resources globally. In Malaysia, the highest production was coming from the state of Johor. Some hitches confronted by the green mussel culturists especially with seafood safety issues. Mollusc diseases not only cause a sense of insecurity in food resources but also the source of monetary loss to the particular culturists. Likewise, in fish culture, parasite infection also one of the major issue in bivalves' culture. The study was conducted on the west coast of Johor where green mussels from Pontian Besar, Pontian, Sungai Lurus, Batu Pahat and Pasir Gudang, Johor Bahru were analysed and it shown to have infestation of few species of parasites such as *caligus sp*, *nematode sp* and *macrogamont-like coccidian*. Somehow samples from Kesang, Muar was not infected with any parasite.

Keywords: Green mussel; *Perna viridis*; parasite; mussels' diseases

Short biography

Mohd Lazim bin Mohd Saif is currently working as Research Officer at the Fisheries Research Institute Gelang Patah, Johor, Malaysia. He completed his Bachelor of Science in Ecology from the Universiti Malaya. He has 10 years experience in aquaculture and havr authored several publications in various journals and books. His publications reflect his research interests in Aquaculture and Fisheries Resources.

Characterisation of pathogenic *Aeromonas* species from diseased tilapia and catfish farmed in East Nusa Tenggara, Indonesia

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Indonesia is one of the largest aquaculture producers worldwide, and like most intensive aquaculture systems, suffers from animal losses due to infectious disease outbreaks. This present study aimed to characterize the motile *Aeromonas* species recovered from natural disease outbreaks occurring in tilapia and catfish (*Clarias spp*). A combination of phenotypic and genomic methods was applied, where a total of 40 bacterial isolates recovered from moribund fish belonged to motile *Aeromonas* species with the highest number of strains identified as *A. veronii* (n=22), *A. hydrophila* (n=15), *A. caviae* (n=1), and other *Aeromonas sp* (n=2). Of the 12 virulence genes 95% of all *Aeromonas* species contained the cytotoxic enterotoxin (*act*) gene. Whereas the aerolysin (*aerA*) gene was presented by 55% of the strains. The highest number of virulence genes and phenotypic expression of the virulence properties was found in strain *A. hydrophila* 92HK with 10/12 genes and 6/6 phenotypic expression. All of the strains were sensitive to sulfamethoxazole (25 µg), enrofloxacin (5 µg), and 100% resistant to amoxicillin (10 µg) with 14/40 of the *Aeromonas* strains were resistant to oxytetracycline (30 µg), of which 13/14 had *tetE* gene. These finding provides critical information on the characteristics of the pathogenic motile *Aeromonas* species associated with infectious disease outbreaks in these systems in Indonesia.

Keywords: fish disease, bacterial infection, *Aeromonas*, virulence profiles, antibiotics resistance.

Bacterial microbiota of Freshwater Prawn, *Macrobrachium rosenbergii* (de Man, 1879) cultured in cages and ponds in the Philippines

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The bacterial quantity and species composition of cage- and pond-cultured freshwater prawn (*Macrobrachium rosenbergii*) were examined over five sampling periods, i.e., at 15-day interval over a period of 75 days after stocking for grow-out culture in floating net-cages in a lake and in earthen ponds, along with the important water physicochemical parameters. The levels of water physicochemical parameters did not show any significant variations throughout the sampling period except for dissolved oxygen but was within the normal range required for the grow-out culture of freshwater prawn. Aerobic plate counts (APCs) of the culture water ranged from $4.7 \pm 1.6 \times 10^6$ to $3.1 \pm 1.2 \times 10^8$ CFU mL⁻¹ and $2.1 \pm 0.2 \times 10^7$ to $8.8 \pm 0.6 \times 10^7$ CFU mL⁻¹ for cage and pond, respectively; whereas APCs in the hepatopancreas of post larvae ranged from $1.8 \pm 0.5 \times 10^5$ to $7.0 \pm 0.1 \times 10^7$ CFU g⁻¹ and $2.3 \pm 0.7 \times 10^6$ to $3.8 \pm 0.4 \times 10^8$ CFU g⁻¹ for cage and pond-reared *M. rosenbergii*, respectively. In the pond sediments, APCs ranged from $7.4 \pm 0.1 \times 10^7$ to $8.0 \pm 0.2 \times 10^8$ CFU g⁻¹. In terms of composition, Gram-negative bacteria predominantly accounted 75% of the total isolated strains with 10 bacterial genera and 12 species, and 9 genera and 13 species identified among cage- and pond-reared *M. rosenbergii*, respectively. *Aeromonas hydrophila*, *A. salmonicida* and *Plesiomonas shigelloides*, and *Chryseobacterium indologenes*, *Enterobacter cloacae* and *Acinetobacter lwoffii* dominantly constituted the bacterial microbiota of pond- and cage-reared freshwater prawn, respectively. Since all *M. rosenbergii* PL samples collected during the sampling appeared to be apparently healthy, along with the fact that no mortalities were recorded, it can be deduced that APCs of $\leq 10^8$ CFU mL⁻¹ in culture water and APCs of $\leq 10^8$ CFU g⁻¹ in PL and pond sediment could be considered as putative threshold values that may serve as baseline information for aquaculturists actively engaging in *M. rosenbergii* culture in the Philippines. Generally, the commensal bacteria identified in the culture water and sediment were also present in the hepatopancreas of *M. rosenbergii* including opportunistic pathogens which under conditions of stress may inadvertently lead to disease epizootics. Thus, good farm management practices need to be adopted to improve the hygienic status of farm-reared prawns.

Keywords: Microbiota, Qualitative, Quantitative, Bacteria, *Macrobrachium rosenbergii*

Short Biography

Dr. Camarin is an Assoc. Professor and Dean at the College of Fisheries and Aquatic Sciences, Mindanao State University. She received her PhD in Fisheries from the University of the Philippines-Visayas after completing her MSc in Aquaculture from the University of Ghent, Belgium. She is also an active associate member of the National Research Council of the Philippines.

Occurrence of Tilapia lake virus (TiLV) in tilapia and tinfoil barbs at wild and culture sites in Malaysia

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Tilapia lake virus (TiLV) is one of the freshwater fish's emerging viral diseases, especially in tilapia. TiLV or *tilapia tilapinevirus* infects all stages of fish from fry to adult and causes massive mortalities in wild and cultured tilapia. Lately, TiLV has been reported to affect other freshwater fish species either through the experimental challenge or in wild. This paper will present the current findings of TiLV research done during the period of 2018 – 2020. A periodic sampling was conducted at four chosen sites with various background, and the species sampled were tilapia and tinfoil barb. The objective is to determine the prevalence of TiLV at all these sites and species. The type of samples taken were organs at 3 of the sites and blood from broodstocks at one site. The organs samples were subjected to cell culture inoculation and reverse transcriptase polymerase chain reaction (RT-PCR) test. In contrast, the potential use of blood samples in detecting the virus was investigated using RT-PCR. In-house primers based on segment 3 of TiLV were designed and used throughout the RT-PCR tests. Results revealed a medium to high prevalence of TiLV in both species at all sites except at 1 hatchery. The TiLV was also consistently found following an outbreak case previously encountered. The TCID₅₀ showed that a higher virus titer was found in tinfoil barb compared to tilapia but at a slower rate of CPE formation. A phylogenetic tree analysis showed that all TiLV isolated from this study had at least 98% similarity with TiLV isolate from Thailand (KY381578.1), indicating that the strain of TiLV had the closest kinship. This project confirmed the prolonged positive detection of TiLV in tilapia and tinfoil barb once an outbreak occurred. From our observation, the affected hatchery will take an extended recovery period as most of the screened stocks were positive. This result alerts the industry to put extra effort into preventing and mitigating the virus in both tilapia and tinfoil barbs.

Keywords: Tilapia Lake virus, TiLV, Malaysia, tilapia, tinfoil barbs

Short Biography

Dr Azila is currently a senior research officer at national fish health research division, DOF Malaysia for 22 years. She obtained her doctor of veterinary medicine and master science degree from university putra Malaysia and has experienced working as a research scientist at the Norwegian veterinary school for 4 years. Her research interest is in the fish virology and currently working on the epidemiology and vaccine development of tilapia lake virus and viral nervous necrosis respectively. She has been authored several publications in various local and impact journal and presenting in numerous local and international seminars, mostly on the field of fish virology.

In-Vitro* antimicrobial activity of probiotics incorporated with micro feed against pathogenic *Vibrio parahaemolyticus

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Micro feed is one of the innovative ways to provide beneficial effects in the early stage of aquaculture species. Incorporation of micro feed with probiotics can potentially enhance the beneficial performance of feed in the host. Probiotics are widely known for their beneficial impacts on gut maturation, growth improvement, immune response and survival rate in most aquaculture species. Hence, the objective of this study was to determine the compatibility and antimicrobial activity of probiotic strains that are to be incorporated into micro feed prototype. A total of 12 potential probiotics used in this study were previously isolated from various hosts and have shown antagonistic properties against multiple pathogens. The potential probiotics were identified as L9 and L11 (*Bacillus amyloliquefaciens*), LAB 3 and LAB 4 (*Enterococcus hirae*), SPS 11, A1, A2 and CL3 (*Lysinibacillus fusiformis*), NAS32 (*Lysinibacillus sphaericus*), I24 (*Bacillus megaterium*), S2 (*Bacillus pocheonensis*) and G87 (*Enterobacter ludwigii*). Spot and well diffusion assays were conducted to evaluate the compatibility between potential probiotics and micro feed. Co-culture assay of probiotics against *Vibrio parahaemolyticus* (10^6 CFU mL⁻¹) was also conducted. All the potential probiotics except for CL3 were compatible with micro feed. Potential probiotics which were compatible with the micro feed were further tested using similar *in vitro* assays to determine the antagonistic properties against pathogenic *V. parahaemolyticus*. Among all the potential probiotics tested, *B. amyloliquefaciens* L9 and *L. fusiformis* A2 demonstrated inhibition zones in both spot and well diffusion assays which indicate antagonistic activity against *V. parahaemolyticus*. The inhibitory zone produced by *B. amyloliquefaciens*, L9 were 0.58 ± 0.01 mm and 0.47 ± 0.01 mm. Meanwhile, *L. fusiformis*, A2 recorded an inhibitory zone of 0.55 ± 0.05 mm and 0.50 ± 0.03 mm respectively in both assays. On the other hand, both strain L9 and A2 at the concentration of 10^6 and 10^8 reduced the growth of *V. parahaemolyticus* (10^6 CFU mL⁻¹) in co-culture assay ($p < 0.05$). Based on the compatibility, *in vitro* screening, and co-culture assays, *B. amyloliquefaciens*, L9 and *L. fusiformis*, A2 could be potentially incorporated into the micro feed to serve as a feed with a disease control properties against *V. parahaemolyticus* infection.

Keywords: probiotics, micro feed, antimicrobial, co-culture, *Vibrio parahaemolyticus*

Infectivity and genome persistence of *Tilapia tilapinevirus* in water samples

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Tilapia tilapinevirus or Tilapia lake virus (TiLV) is a high contagion pathogen that affects global tilapia aquaculture. To date, TiLV continues to threaten tilapia production and food security as shown by the detection of the virus in 16 countries across four continents. Previously, studies showed that TiLV spread in fish vertically and horizontally by infecting fish, eggs, and broodstock. Nevertheless, the persistence of the virus in contaminated water, materials, or potential carriers has not yet been investigated. In this study, we evaluated the persistence and infectivity of TiLV in three different types of water: sterile distilled water (SDW), freshwater collected from rearing fish tanks (FW), and natural pond water (PW). The virus was spiked with the stock virus at 0.6 mL of 3.18×10^7 viral copies/mL of water and then incubated at 27°C for 0, 3, 5, 7, 10, and 14 days post-inoculation. Samples were concentrated and filtered through an electronegative charge membrane to collect virus RNA and evaluated by real-time PCR. At 14 days, TiLV genome in SDW was 5.09 log₁₀ reduction, while the virus genome is detected in FW and PW up to 3 and 7 days, with most of the virus already disappear (reduction above 4 log₁₀). Further infectivity study revealed that the virus lost its infectivity within 1 day in all three water samples, despite it having been spiked at 10 times higher than the concentration used in the persistence study. Collectively, this new information provides the first evidence to support that TiLV may persist and cause infection in fish rearing water. Appropriate control strategies including disinfecting or resting reservoir water for at least three to five days prior to stocking tilapia should be applied in the farm biosecurity practice to limit the spread of TiLV.

Keywords: Persistent, *Tilapia tilapinevirus*, infectivity, concentration, water sample

Short Biography

Jidapa Yamkasem is a Ph.D. student in the Graduate Program in Animal Health and Biomedical Science, Faculty of Veterinary Medicine, Kasetsart University, Thailand. Her research focuses on emerging viral diseases that affect tilapia production.

Global seahorses diseases: A Review

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Seahorses are facing extinction under the increasing pressure of human activities and climate change worldwide. The biggest threat that seahorses face comes from collection from the wild for the marine ornamental trade, traditional Chinese medicine (TCM), and curios. Aquaculture as an alternative to the problems described requires major advances in culture techniques and maintenance. Currently, the majority of seahorse aquaculture involves small-scale operations, especially in developing countries. Disease and feeding were identified to be the most pressing concerns in seahorse aquaculture. This review was on the historical and current information on global seahorse diseases, including species in culture and technical issues when raising seahorses in captivity. This paper brings together scientific data that will help us understand the health of natural and cultural seahorses. It also contributes to the development of efficient strategies for disease management in aquaculture.

Keywords: Seahorses, diseases, sustainable aquaculture, knowledge gap, aquaculture

Short biography

Marjorie Charam is currently working as a Research Officer at the Fisheries Research Institute (FRI). She received her Master's degree by Research in Aquaculture (Marine Biotechnology) from the University of Malaysia Terengganu (UMT), Terengganu. Her research interests and publications are related to marine biotechnology and ornamental aquaculture, particularly marine ornamental aquaculture.

Genomes and virulence factors of novel *Pseudomonas koreensis* strain isolated from aquaculture farm in Kuching, Sarawak, Malaysian Borneo

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Pseudomonas koreensis have been reported as pathogens in freshwater fish. Unlike environmental *P. koreensis* from soil, which has been studied extensively for its role in promoting plant growth, pathogenic *P. koreensis* from fish has been assessed only to a very limited extent. This study is, to the best of our knowledge, the first report on the complete genome sequences of the novel pathogenic strain, *P. koreensis* ML-1 isolated from the gut of diseased *Tor tambroides*, and the potential virulence factors used by this pathogen to infect fish. We achieved a high-quality genome of *P. koreensis* ML-1 with a N50 value of 233,601 bp and completeness of 99.5% according to BUSCO. The total size of the *P. koreensis* ML-1 genome contains 6,171,880 bp and has a G+C content of 60.5%. The genome sequence includes 5547 protein-coding genes, 3 rRNA genes, 65 tRNAs, and no plasmids were found. Besides these, the prediction of virulence genes was performed using the VFAnalyzer software from the Virulence Factor Database (VFDB). A total of 17 virulence classes which consisted of 64 virulence factors and 149 virulence genes were identified in the genome of *P. koreensis*. Genes involved in adherence were among the most prevalent (n=78), followed by those involved in anti-phagocytosis (n=24), and iron uptake (n=14). Although the functions of these virulence factors need yet to be deciphered experimentally, the availability of this genome provides the basic understanding of the pathogenesis of *P. koreensis* and emphasizes the need for further investigation of these mechanisms.

Keywords: *Pseudomonas koreensis*; virulence factors

DAY 2: 24 August 2022 (Wednesday)

**Session 3: Detection Method/Diagnostic
(Parasitic, Bacterial & Viral Diseases)**



Plenary II

State of global aquaculture

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According to FAO, in 2020, global fisheries and aquaculture production reached a record 214 million tonnes, comprising 178 million tonnes of aquatic animals and 36 million tonnes of algae (seaweed), largely due to the growth of aquaculture in Asia. The global aquaculture production in 2020 was also a record 122.6 million tonnes, with a total value of USD 281.5 billion. Aquatic animals accounted for 87.5 million tonnes and seaweed comprised 35.1 million tonnes. However, aquaculture growth in Africa records a reduction due to a decrease in the production in two major producing countries, Egypt, and Nigeria. In terms of consumption, current annual per capita consumption of fish in Africa is less than 10kg (global average is 20.2 kg) and is alarmingly forecasted to reduce in the coming years. However, rising incomes and urbanization, improvements in post-harvest practices and changes in dietary trends are projected to drive a 15 percent increase in aquatic food consumption, globally to supply on average 21.4 kg per capita in 2030. Whether this increase will have any impact on Africa is questionable. This presentation explains multiple facets of the state of aquaculture, their prospects, bottlenecks, strategies and solutions at global, regional and national levels.

Keywords: Aquaculture, food consumption, production, global, growth

Short Biography

Dr Rohana Subasinghe is a specialist in aquaculture development and aquatic animal health management. After 22 years of service, he retired from FAO in October 2015. He was responsible for the implementation of many programmes and projects on aquaculture and aquatic animal health at national, regional, and international levels, worldwide. For fifteen years, he served as the Technical Secretary to the Sub-Committee on Aquaculture of the Committee on Fisheries of the FAO. A former teacher at the University of Colombo and the Universiti Putra Malaysia, Dr R.Subasinghe earned his PhD from Stirling University, Scotland. He is a former Chairperson of the Fish Health Section of the Asian Fisheries Society; 2018-2019 President of the Asia Pacific Chapter of the World Aquaculture Society (WAS-APC); and an honorary life member of the World Aquaculture Society and Asian Fisheries Society. He is a strong advocate for the need for more inclusive growth of global aquaculture. Dr. R.Subasinghe is currently working with World Fish, Bill & Melinda Gates Foundation and the United States Agency for International Development, towards improving fish supplies in Africa through aquaculture.

Keynote 3

Fish wear their immune system on the outside – what this means for aquaculture and ecology

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After 40 years of intensive aquaculture, we know the fish as an *animal* better than we ever did from millenia of fishing. The fundamental differences between aquaculture and agriculture animals include the time spent in a controlled stable environment for organogenesis (upto 70% of total lifespan for terrestrial's vs 0-5% in aquatics), the number of evolutionary whole genome duplication events contributing to the current genome (2R for terrestrials and 3R or 4R for aquatics) and the body location of the mucosal immune system (inside for terrestrials and both inside and outside for aquatics). The external immune system of the slimy layers (mucosa of skin, gills and intestines) contains antiviral, antifungal, antiparasitic and antibacterial substances in constant dialogue with the environment. The mucous cells exhibit an organ-wide repeatable response to stimuli such as therapeutics, stress, diet and environment. The application of an unbiased standard method, mucosal mapping or Veribarr™, over 12 years and over 100 trials in ecology and aquaculture has given unprecedented understanding of how teleost mucosa function. Gills, which comprise about 50% of the surface area of fish, have proven to be the most sensitive early warning of systemic dysregulation. Some results from commercial-scale productions, from “detective work” and from controlled lab studies will be highlighted in the talk along with a plea for development of standards for fish health. The scope for growth in aquaculture resides in *maintaining* good stock health.

Short Biography

Pittman is a tenured professor in the Fisheries Ecology and Aquaculture Research Group, Department of Biology, at the University of Bergen, Norway. She is a Canadian educator, inventor, entrepreneur and scientific consultant with an international background. Working in Norway since the 1980s, Prof. Karin Pittman is a socially engaged researcher who has contributed within fisheries research in developing countries, aquaculture research, business development, teaching and diplomacy, as Honorary Consul for Canada 1999-2015. As a researcher, she has studied egg and larva development in farmed marine species, nutrient uptake and food webs, marine ecology and fish health aspects. Her studies of the importance of the slime layer for health led to the establishment of Quantidoc AS in 2013, as well as an Inventor Award from Hordaland County Council in 2013 and the Global Aquaculture Alliance Prize for Aquaculture Innovation and Leadership in 2016 based on Mucosal Mapping now trademarked as Veribarr™. Karin Pittman is also a committed and creative educator who has received several awards for her teaching,

including Olav Thon's Award for Outstanding Teaching in 2016, the Lecturer Prize at the Department of Biology 2012, the Owl Prize (UiB) and the NOKUT Award for Best Course in Higher Education in 2009. She has served with various international Research Council boards and programmes. She is a member of the Consular Corps Norway. She is also a grateful alumnus of the 2012 Senior Workshop at the "Alan Alda Center for Communicating Science" at Storybrook University of New York.

Oral Presentation

Profile of protein Isolated from mucus of infected *Oreochromis* spp.

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Streptococcus agalactiae is an emerging pathogen which causes a severe systemic disease in fish worldwide. Fish epidermal mucus contains innate immune components that provide the primary defense against different pathogenic microbes. Quick detection of Streptococcosis under field conditions is very helpful in determining if prophylaxis is needed. To investigate this issue, 30 red tilapia *Oreochromis* spp. each of 150g were infected with *Streptococcus agalactiae*, *Staphylococcus aureus* and *Aeromonas hydrophila*. Every 1, 2, 4, and 8 hours, fish body mucus was collected in order to profile and explore molecular changes after subsequent challenge towards the causative agent. Sodium Dodecyl Sulfate Polyacrylamide Gel-Electrophoresis (SDS PAGE) were used to allow the fish body mucus protein separation by mass and western immunodetection using specific polypeptides were used to identify the immunogenic protein. As a result, common protein, 14 kDa was found in all of the isolated mucus challenged. Meanwhile, protein with size 49 kDa, 81 kDa and 101 kDa were found as prominent protein of *Streptococcus agalactiae*. The prominent protein after challenge with *Staphylococcus aureus* are 20 kDa, 30 kDa, 35 kDa and 63 kDa. While the most prominent protein after challenge with *Aeromonas hydrophila* are 35 kDa, 40 kDa, 60 kDa. All of this finding is important towards a better treatment and prevention of disease occurrence in Tilapia aquaculture.

Keywords: Freshwater; protein profiling; *Streptococcus agalactiae*; Tilapia

Short Biography

Muhamad Faizal bin Mohd was born in Kajang, Selangor, on October 3, 1994. I completed my Bachelor of Science (Marine Science and Technology) from the International Islamic University Malaysia, Kuantan, Pahang (2018), currently pursuing Master in Science (Biotechnology) in International Islamic University Malaysia, Kuantan, Pahang. Working at the National Fish Health Research Center as Research Assistant. Has been involved in research activities at the National Fish Health Research Centre (2018 and 2022). Has published in the International Journal of Agriculture, Environment and Biotechnology

Rapid CRISPR-based platforms for detection of major pathogens in penaeid shrimp and fish

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Infectious diseases in cultivated shrimp and fish can result in rapid mortality, growth retardation, and compromised immune systems, leading to dramatic economic losses in the global aquaculture sector. While a variety of molecular diagnostic techniques are currently available for disease screening and surveillance, they are constrained by their long turnaround time, requirement of trained operators, and demand for sophisticated instrumentation. To address this critical gap, our research group has successfully developed CRISPR-based assays for the detection of major diseases in shrimp and fish, including white spot disease virus (WSSV), *Enterocytozoon hepatopenaei* (EHP), scale drop disease virus (SDDV), and tilapia lake virus (TiLV). The CRISPR-based detection method is centered on an RNA-guided endonuclease called Cas12a, which generates visual signal in the presence of nucleic acid sequences matching the so-called guide RNA. These assays, when combined with recombinase polymerase amplification, enable the detection of as few as 50 copies of target per reaction and exhibit no cross-reactivity with other closely related pathogens. Moreover, the method is isothermal (37 – 42 °C), takes only an hour from sample to answer, and provides facile visual readouts via lateral flow detection and portable fluorescence device. Based on these advantages, the CRISPR platforms should be an attractive alternative that is highly suitable for in-field implementation while offering levels of sensitivity and specificity comparable to, if not exceeding, those of conventional molecular techniques.

Keywords: Clustered regularly interspaced short palindromic repeats (CRISPR), isothermal detection, white spot syndrome virus, EHP, scale drop disease virus

Short Biography

Dr. Thawatchai Chaijarasphong is currently working as an assistant professor at Centex Shrimp and the Department of Biotechnology at Mahidol University, Thailand. He received his PhD in Chemistry (Chemical Biology) from the University of California, Berkeley. Dr. Chaijarasphong has authored several publications in various journals, and his publications reflect his research interests in molecular diagnostics and CRISPR applications in aquaculture.

Application of a specific antibody to localize tilapia lake virus (TiLV) in fish tissues using Immunohistochemistry

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Tilapia lake virus (TiLV) or *Tilapia tilapinevirus* is a novel RNA virus in tilapia causing high economic risk to the global tilapia aquaculture. In this study, we generated a rabbit polyclonal immunoglobulin G antibody specific against TiLV and applied it for a new immunohistochemical (IHC) protocol. Our results revealed that TiLV protein was detected in the nucleus and cytoplasm in cells of different organs, which is in accordance with *in situ* hybridization (ISH) findings. Specifically, TiLV IHC signals were detected in the areas of tissue inflammation, circulating leukocytes in the blood vessels, and endothelial cells of various organs, suggesting the cellular tropisms of the virus. Our data displayed the TiLV distribution pattern in infected fish and suggested that this virus has endotheliotropism and lymphotropism, which requires further investigation. Generally, TiLV positive staining was commonly found in intestines, gills, and brain, indicating that these organs are the primary targets for TiLV. Further analysis using the new antibody and IHC technique to detect TiLV infections in various fish species including ornamental cichlids, Mozambique tilapia, giant gourami, and naturally infected tilapia supported the application of IHC to localize TiLV. Taken together, the developed IHC platform can be applied to understand the pathobiology and virus-host interaction during TiLV infection.

Keywords: Immunoglobulin, Immunohistochemistry, *In situ* hybridization, *Tilapia tilapinevirus*, Cellular tropism, diagnosis

Short Biography

Puntanat Tattiyapong is a PhD candidate in Interdisciplinary Program in Genetic Engineering and Bioinformatics, Graduate school, Kasetsart University. Her research interest is on the development of Tilapia lake virus vaccine.

An intelligent protozoan white spot fish disease detection

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Protozoan white spot disease is one of the leading causes of fish disease outbreaks among mariculture fish populations in Malaysia. The outbreak induced by an active parasite, *Cryptocaryon irritans* is hazardous as it could impair the production and cultivation of diverse species resulting in a negative impact on the expansion of the aquaculture industry. As part of the emergence of Industrial Revolution 4.0, maximizing the use of Artificial Intelligence in the industry could be the key contributor as current technologies in disease detection could contain or slow the spread of infections among cultured fish. The advances of image processing in the underwater environment and deep learning techniques prove it is possible to reduce the need for human observers in fish disease detection. Hence, this study demonstrates a new procedure for white spot disease detection using underwater images, concerning on the development of integration between contrast-limited adaptive histogram equalization (CLAHE) with convolutional neural network (CNN). A total of 40 validated normal and infected underwater fish images have been tested in this study. The techniques used has achieved 94.67% average accuracy of white spot disease detection. Therefore, an intelligent protozoan white spot fish disease detection could be serve as an alternative to an early detection of the disease, thus preventing from secondary infection in mariculture fishes. The incentive offered could ensure the anticipated growth of the industry as the sector plays a significant source of economic income for both country and farmers.

Keywords: Protozoan White Spot, Mariculture, Disease Detection, Contrast-Limited Adaptive Histogram Equalization, Convolutional Neural Network

Short Biography

Siti Naquiah Md Pauzi and Amiera Syazlin Md Azhar are currently studying in Master of Science (Technology Management) and Master of Science (Intelligent System) respectively at Universiti Utara Malaysia. Both authors are supervised by Mohamad Ghozali Hassan, Nor Hazlyna Binti Harun, Noraini Yusoff and Kua Beng Chu; senior lecturers and researchers from Universiti Utara Malaysia, Universiti Malaysia Kelantan and National Fish Health Research Centre. The authors have a growing interest in the fields related to image processing and Artificial Intelligence.

Gut metagenome analyses of healthy and diseased tilapia *Oreochromis niloticus* cultured in Bangladesh

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Gut microbiota plays an indispensable role in fish fitness or health. Intestinal disorders regulated with the composition of microorganisms which act as a barrier against pathogens. This study was focused on the composition of intestinal microbiota between the healthy (HF) and diseased (DG) Nile tilapia through NGS based 16S rRNA sequencing. For this purpose, two HF and two DG Nile tilapia (fishes with symptoms like swollen anus, exophthalmia, inflammation and abdominal distension) with replicates were collected from Narsingdi and Chandpur of Bangladesh. A total of 41 phyla with 548 genera of bacteria were found in healthy tilapia and 46 bacterial phyla with 600 genera were identified in diseased tilapia. This study revealed that the dominant phylum was Verrucomicrobia in healthy- and Chloroflexi in diseased-Narsingdi samples. On the other hand, both healthy and diseased-Chandpur tilapia contained the highest reads of Proteobacteria. At the genus level, highest abundance observed for *Rubritalea* (healthy-Narsingdi tilapia) and *Thermogemmatispora* (diseased-Narsingdi tilapia) whereas healthy and diseased tilapia of Chandpur contained *Bifidobacterium* and *Thermogemmatispora* genera most abundantly. 5.39% and 9.68% of phyla were unclassified in healthy and diseased Narsingdi samples whereas 8.84% and 9.08% reads of healthy and diseased Chandpur samples were unclassified phyla. Several pathogenic and beneficial genera were identified *viz.* *Vibrio*, *Staphylococcus*, *Clostridium*, *Bacillus*, *Streptococcus*, *Enterobacter*, *Mycobacterium*, *Pseudomonas*, *Enterococcus* from the studied samples. Overall, relative abundance of Proteobacteria, Chloroflexi, Actinobacteria, Firmicutes were increased in diseased tilapia than healthy sample. Besides, healthy fish contained greater reads of Verrucomicrobia, Fusobacteria than diseased fish. Findings of this study revealed the changes in the composition of gut microbiota of healthy and diseased tilapia. This research could be helpful to figure out future investigation for preventing and controlling disease infected tilapia.

Keywords: Gut microbiota, *Oreochromis niloticus*, 16S rRNA gene, NGS, Bangladesh

Short Biography

Dr. Mohammad Shamsur Rahman is currently working as Professor of Aquaculture at the Department of Fisheries, University of Dhaka, Bangladesh. He received PhD on Veterinary Sciences from the University of Padova, Italy. He completed Masters in Fisheries from the University of Dhaka. He then worked at ICDDR,B, NSTU and Dhaka University. He has authored more than thirty publications in various journals and books. He has supervised more than thirty MS students and supervising three PhD researchers. His publications reflect his research interests in Aquaculture and Health Management. He is running a small group especially dedicated to Aquatic Animal Health Research.

Glucose and glutamine contributed to *de novo* nucleotide synthesis to support viral pathogenesis

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To support virus replication, the host metabolic system is typically hijacked and rerouted to create an appropriate cellular environment with an abundance of metabolites. Previous reports indicated that WSSV facilitates aerobic glycolysis (Warburg effect), with a high rate of glucose oxidation. Based on this hypothesis, we verified that metabolites in the pentose phosphate pathway and nucleotide synthesis significantly accumulated during the WSSV replication stage. Furthermore, glutamine is perceived as the crucial metabolite contributing to viral replication. However, the mechanism of how WSSV regulates host nucleotide and glutamine metabolism is not well understood. The objective was to determine involvement and contributions of *de novo* nucleotide biosynthesis during WSSV replication. First, we used LC-ESI-MS and isotopically labelled glucose ([U-¹³C] glucose) and glutamine ([A-¹⁵N] glutamine) as metabolic tracers and confirmed that *de novo* nucleotide synthesis pathways were activated at the replication stage (12 h-post infection, 12 hpi). Next, we confirmed several genes involved in nucleotide synthesis were significantly upregulated at 12 hpi. Then, in shrimp treated with specific dsRNAs to knockdown genes and subjected to WSSV infection, there were significant decreases in both WSSV gene expression and genome copy numbers. Therefore, we inferred that *de novo* nucleotide synthesis was involved in WSSV pathogenesis. Nevertheless, how viral protein modulated and interacted with host protein involved in nucleotide synthesis still needs to be investigated. Taken together, this research not only clarified that host nucleotide synthesis had a crucial role in WSSV pathogenesis, it has also provided new knowledge for evidence-based approaches to control WSSV outbreaks.

Keywords: White spot syndrome virus (WSSV), Warburg effect, Nucleotide synthesis

Short Biography

Mr. Cong-Yan Chen is currently working as a PhD student at the National Cheng Kung University. He received his BSc degree in 2020 from National University of Kaohsiung. His research is focused on how white spot syndrome virus (WSSV) modulates and facilitates *de novo* nucleotide synthesis in shrimp to promote viral pathogenesis.

3-Minutes Presentation

Mass mortalities of golden pomfret (*Trichinotus blochii*) at floating cages Pulau Aman, Penang associated with oxygen crisis, multiple infections of parasites and *Vibriosis*

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Mass mortalities of golden pomfret aged between 1-5 months old have been reported by fish farmers at floating cages offshore near Pulau Aman, Penang on 20th September 2020. Golden pomfret aged between 1-2 months old were badly affected with 80-100% mortalities. Hence, the objective of this study is to determine the cause of fish mortalities occurring at the cage culture farms, Pulau Aman, Penang. On site investigation with sample of fishes (n=13) and waters from the affected cages were taken for laboratory analysis. Parasite was examined from skin and gills, aseptic method for bacterial inoculation and isolation on tyryptose soy agar (TSA+1.5% NaCl) were performed from the internal organ of kidney, spleen and liver. Pooled organs tissues were preserved in 95% alcohol for detection of Virus Nervous Necrosis (VNN) and Iridovirus (RSIVD) using polymerase chain reaction. *In-situ* analysis of water parameters (YSI 5908 probe, USA) showed low dissolved oxygen (3.0-3.5 mgL⁻¹) while other parameters were within positive range for aquaculture. High prevalence of *Neobenedenia* sp. (92%) and marine leech *Zeylacobdinella arugamensis* (31%) were detected in golden pomfret with mean intensity of 5 and 2, respectively, as the fish have been treated with fresh water bath a day earlier. Secondary infections of *Vibrio vulnificus* (69%) and *V. alginolyticus* (31%) were found as mixed of two isolates or a single pathogen in either one or more organs of sick fish. We believed, high mortalities of golden pomfrets occurring on 20-22 September 2020 were caused by multiple factors mainly low dissolved oxygen, parasitic infestations of capsalid *Neobenedenia* sp., marine leech and *Vibriosis*. The affected fish were found with lesions such as scale drops, fin and tail rot, skin wound from mild to severe ulceration and haemorrhages on body due to co-infection of parasites and *Vibriosis*.

Keywords: mass mortalities, low dissolved oxygen, parasites, *Vibriosis*

Short Biography

Dr Padilah Bakar is currently a Research Officer at the NaFisH, FRI Batu MAUNG. She completed her Doctor of Veterinary Medicine from the Universiti Putra Malaysia (UPM) and MSc from Universiti Sains Malaysia. Her research interest is in bacterial diseases of fish and shrimps.

Experimental co-infection of tilapia lake virus and *Streptococcus agalactiae* in red hybrid tilapia (*Oreochromis niloticus* × *O. mossambicus*) model

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Tilapia tilapine virus or better known as Tilapia Lake Virus (TiLV) is an emerging virus accountable for a viral disease in tilapia. It has been responsible for a massive mortality of tilapia around the globe including Malaysia. Recently, few studies have found a concurrent infection between TiLV and other pathogens including *Streptococcus agalactiae* in tilapia which contributed to a high death rate. This study determined the relationship of TiLV and *S. agalactiae* co-infection in red hybrid tilapia (*Oreochromis niloticus* × *O. mossambicus*) following intraperitoneal (IP) injection route. The red hybrid tilapias were challenged with different concentrations of TiLV and *S. agalactiae*. Following the infections, the LD_{50-20dpi} of TiLV and *S. agalactiae* was successfully determined at 10⁶ TCID₅₀/mL and 10⁴ CFU/mL. The clinical signs, gross lesions, and histopathological changes of TiLV and *S. agalactiae* challenged fish were similar as observed in the naturally infected fish. Afterwards, by referring to the LD_{50-20dpi}, the fish were challenged with 10⁶ TCID₅₀/mL of TiLV and 10³ CFU/mL of *S. agalactiae* following the single and concurrent infection. The co-infected challenged fish showed a high cumulative mortality (60.00% in TiLV-*S. agalactiae* co-infection, and 73.33% in *S. agalactiae*-TiLV co-infection) as compared to the single challenged fish (40.00% in TiLV infection and 20.00% in *S. agalactiae* infection). Serious histopathological findings were also found in the co-challenged fish as compared to the single challenged fish. Analysis of viral and bacterial load recovered from the fish's organs (brain and liver) in qPCR analysis showing a higher load pattern were observed in the co-challenged fish. The virus and bacteria were also observed using the TEM analysis and the analysis has confirmed the presence of TiLV and *S. agalactiae* in the challenged fish. Our study highlights the possibility of co-infection between TiLV and bacterial pathogen as a serious threat towards the Malaysia's tilapia industry.

Keywords: Tilapia Lake Virus, *Streptococcus agalactiae*, clinical signs, histopathological changes, qPCR, TEM

Short Biography

Lukman Basri is currently working on his Master of Science (Aquatic Biotechnology) at Institute of Bioscience, Universiti Putra Malaysia. He received his Bachelor Degree on Biology from Universiti Putra Malaysia. He is currently focusing on virology and bacteriology infection in fish. Lukman has co-authored several publications in various journal. His publications reflect his research interests in fish virology and bacteriology.

Investigation of culture condition inducing viable but non-culturable (VBNC) state in *Nocardia seriolae*

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Nocardiosis caused by *Nocardia seriolae* occurs in marine aquaculture of amberjack species. The main symptoms of the disease are nodule formations in the gills, spleen and kidney. In previous studies, the bacteria were detected in the spleen of outwardly healthy farmed yellowtail (*Seriola quinqueradiata*) by PCR and immunohistochemistry but were not isolated from the tissue on 1% Ogawa medium. These results suggest that the bacteria latently infected in the host spleen as viable but non-culturable (VBNC) state. In this study, we investigated the culture conditions that induce *N. seriolae* to VBNC status. *N. seriolae* precultured in BHI broth were incubated in normal BHI broth, 16-fold diluted BHI and artificial sea water for 45 days with shaking. A portion of the culture were collected periodically and subjected to measurement of OD630 and growth test on 1% Ogawa medium. Further, the portion of the culture were incubated with 5(6)-Carboxyfluorescein diacetate (CFDA) and the viability of the bacteria was determined using flowcytometry. *N. seriolae* cultured in normal BHI broth reached to plateau at 20 days post-incubation but increase of the OD630 values were not observed in the other groups. *N. seriolae* cultured in normal BHI broth for 45 days formed no colony on 1% Ogawa medium and the bacteria cultured in artificial sea water for 45 days formed a few colonies on the slant. However, many colonies were found on the medium inoculated with the bacteria cultured in 16-fold diluted BHI for 45 days. Flowcytometry analyses revealed that 96.4%, 92.1% and 21.6% of the bacteria were positive for CFDA staining in normal BHI broth, 16-fold diluted BHI and artificial sea water, respectively. These results suggest that *N. seriolae* entered VBNC state at 45 days incubation in normal BHI broth with shaking. Moreover, the bacteria cultured in the diluted medium probably fell into dormancy at 45 days incubation.

Keywords : *Nocardia seriolae*, Nocardiosis, VBNC, latent infection, poor nutrition

Short Biography

Mr. Akito Hayashi is currently studying as a master student at the Laboratory of Fish Pathology in Tokyo University of Marine Science and Technology. His research interest is dormancy and VBNC state of fish pathogenic bacteria. He aims to clarify the infection mechanism of nocardiosis through his research and hopes that his research will lead to the development of a vaccine for the disease.

Mucosal responses in giant freshwater prawn, *Macrobrachium rosenbergii* fed with diet containing natural feed premix

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This study was conducted to investigate the mucosal response in giant freshwater prawn (GFP), *Macrobrachium rosenbergii* towards diet mixed with polysulphides derivative premix and its role in enhancing innate immunity and feed conversion ratio (FCR) in prawn. The GFP were fed with diets containing different levels of feed premix :0g/kg (control), 1 g/kg (P1) and 2g/kg (P2). After feeding trial of 60 days, GFP were challenged with *Vibrio parahaemolyticus* and mortality was recorded. GFP's histology samples were collected and a total of 45 tissue sections were stained with Periodic Acid Schiff-Alcian Blue reagent for unequivocal marking of the mucous cells and mounted with paraffin wax. Later, the slides were digitally scanned using MoticEasyScan Pro 6 and saved as high-resolution digital images in jpg-format. Mucosal mapping of the digitized slides was performed using the MucoMaster2 (Quantidoc AS, 2019) software and analysed for mucous cell area and density in GFP's intestine and gills. Significant differences in ($p < 0.05$) in mucous cell area (MCA) and mucous cell density MCD) were recorded between groups fed with different levels of feed premix. The GFP's innate immune system has significant responses to diet and pathogens. These results of this study demonstrated that feed premix might strengthen the non-specific immunity and reduce the mortality in GFP culture.

Keywords: mucosal mapping, *Macrobrachium rosenbergii*, innate immunity, polysulphides, mucous cells

Short Biography

Imelda is currently working as Research Officer at the Fisheries Research Institute, Sarawak and Labuan Division. She completed her Masters in Aquaculture Biology from the University of Bergen, Norway. Her research interest is mucosal immunity.

Experimental challenges studies on pathogenicity of *Edwardsiella ictaluri* in striped catfish, *Pangasianodon hypophthalmus* (Sauvage)

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Striped catfish, *Pangasianodon hypophthalmus* is one of the major species that were being cultured in Malaysia, especially in Pahang. However, the challenge that is being faced by this species is disease due to invasion of pathogen. This study aimed to investigate the pathogenicity and lethal dose (LD₅₀) of *Edwardsiella ictaluri* in *Pangasius hypophthalmus* by mean of injection and immersion. 170 of striped catfish with average weight of 45g ± 2g were used in this study and they were challenged with *Edwardsiella ictaluri* experimentally. 100 of them were immersed and 70 were injected in four different concentrations of *E.ictaluri* and were observed for 20 days straight. The concentration used for injections are 1×10¹⁰, 1×10⁹, 1×10⁸, 1×10⁷ bacterial cells and for immersions are 1 x 10¹¹, 1 x 10¹⁰, 1 x 10⁹, 1 x 10⁸ cfu/ml. Moribund and dead fish were collected for clinical signs observation externally and internally. *P.hypophthalmus* shown the signs of vertical swimming, white spot lesion on internal organ such as liver, formation of ascites fluid all over peritoneal cavity, swollen on the abdomen and abnormal of peritoneal cavity. The highest cumulative mortality was at concentration of 1 x 10¹¹ cfu/ml with 100% mortality and LD₅₀ was 2.03092x10⁹ cfu/ml for immersion. Meanwhile, the greatest mortality for injection method in this study is at 1x10¹⁰ cfu/ml with 100% mortality and LD₅₀ is 8.231x10⁸ cfu/ml. It can be concluded that *E.ictaluri* does affect the survivability of *Pangasianodon* species in Malaysia.

Keywords: Pathogenicity, LD₅₀, *Edwardsiella ictaluri*, *Pangasianodon hypophthalmus*

Short biography

Amira is a full-time master's student in the Department of Marine Science and Technology at International Islamic University Malaysia Kuantan. Her interested field of study is in fish disease. She currently works on epidemiology study of a bacteria called *E.ictaluri* that caused disease in striped catfish, *Pangasius hypophthalmus*.

Pathogenicity and LD₅₀ of *Pangasius nasutus* against *Edwardsiella ictaluri* through injection method

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This study focuses on the pathogenicity and LD₅₀ of *Pangasius nasutus* against *Edwardsiella ictaluri* through injection. Generally, *Pangasius nasutus* is a native freshwater species of peninsular Malaysia and popularly known as Patin Buah among the locals in Malaysia. Most of this species can be found in the rivers of Pahang which in Pekan and Maran districts particularly. The market price for this species is quite high as it tastes better compared to other catfish. Meanwhile, *Edwardsiella ictaluri* is a pathogenic bacteria and causative agent which cause Bacillary Necrosis of Pangasianodon (BNP) and enteric septicaemia of catfish (ESC) in the *Pangasius* species nowadays. However, the prevention measures against the bacteria such as production of vaccine and antibiotic are still unknown for *Pangasius nasutus* species due to the lack of research and information. Therefore, the objective of this study is to determine the pathogenicity and LD₅₀ of *Pangasius nasutus* against *Edwardsiella ictaluri*. There are four different concentrations of *Edwardsiella ictaluri* (1×10^{10} , 1×10^9 , 1×10^8 , 1×10^7) were injected intraperitoneally including normal saline water as control to a total of 50 *Pangasius nasutus*. Observation of clinical signs and mortality were carried out for 28 days and LD₅₀ was determined. The earliest clinical sign was observed at concentration of 1×10^{10} cfu/ml at 2 hours post-infection of *Edwardsiella ictaluri* where pale liver and congested kidney can be seen after dissection. The other clinical signs observed throughout this study are inflammation on tail and fin, hemorrhagic fin, hemorrhagic upper mandible, discoloration, and inflammation on the lower part of the body. The first mortality of *Pangasius nasutus* was at concentration 1×10^{10} cfu/ml at 2 hours of post injection. The highest cumulative mortality was recorded at concentration 1×10^{10} cfu/ml with 100% of mortality rate. From the result, the value of LD₅₀ of *Edwardsiella ictaluri* calculated was 1×10^6 cfu/ml. From this study, it can be concluded that *Edwardsiella ictaluri* does affect the survivability of *Pangasius nasutus* in Malaysia.

Keywords: Pathogenicity; *Pangasius nasutus*; *Edwardsiella ictaluri*; LD₅₀; Catfish

Short Biography

Mohd Syafiq Syauqi bin Mohd Salim was born in Pasir Mas, Kelantan, on March 10, 1997. Completed my Bachelor of Science (Marine Science and Technology) from the International Islamic University Malaysia, Kuantan, Pahang (2021), currently a Graduate Research Assistant (GRA) pursuing in Master Program (Master of Science in Biotechnology) at International Islamic University of Malaysia (IIUM) Kuantan Campus. Has been involved in research activities at IIUM since degree especially during Final Year Project (FYP). Has experience working in aquaculture industry which as Prawn Farm Supervisor at FDI Aquaculture Sdn. Bhd.

***In vivo* expression of acute hepatopancreatic necrosis related virulence genes in *Vibrio parahaemolyticus* and *V. harveyi* and its virulence towards *Penaeus vannamei* (Boone, 1931)**

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Acute hepatopancreatic necrosis disease (AHPND) recently escalates into panzootic, leading to severe losses in shrimp industries. In this study, the expression of AHPND virulence factors, quorum sensing regulator, LuxR and virulence regulator, ToxR in *V. parahaemolyticus* strain BpShHep31 and *V. harveyi* strain BpShHep24, causing AHPND were investigated. A significant ($P < 0.05$) increase in the expression levels of the quorum sensing master regulator *luxR* when compared with the control shrimp (unchallenged group) was detected. There was also a substantial difference in *pirA*, *pirB* and *toxR* expressions in the challenged shrimps compared to unchallenged shrimp. The expression of AHPND virulence genes, *luxR* and *toxR* peaked at 36 h post challenged at 20-30-, 41-59- and 25-39-fold respectively. Furthermore, up-regulation of *pirA*, *pirB*, *luxR* and *toxR* expression reflected in the increasing rate of shrimp challenged with *V. parahaemolyticus* strain BpShHep31 and *V. harveyi* strain BpShHep24. In general, the *in vivo* gene expression study demonstrated a clear difference in term of gene expressed between the challenged (with AHPND positive strains) and unchallenged shrimp.

Keywords: AHPND, *V. parahaemolyticus*, *V. harveyi*, quorum sensing and *in vivo*

Short Biography

Sarmila was a Laboratory Manager in Biovalence since 2014 to Feb 2017. She was leading the Shrimp Disease Centre, under the purview of Blue Archipelago. She has in depth knowledge in scientific reporting, laboratory management and R&D related. She also has successfully mentored and managed over 20 interns from various universities. Prior to Biovalence, she was a Researcher in University of Malaya (UM). Currently, she's pursuing her doctoral degree at Universiti Putra Malaysia (UPM). Her research is more into bacterial disease mechanisms in shrimp and food security which supports the UN-SDG goal number 12 (i.e., Responsible production and consumption).

Phenotypic characterization of *Edwardsiella ictaluri* isolated from cage-cultured *Pangasianodon hypophthalmus*

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Pangasianodon hypophthalmus is the most cultured catfish in Pahang, which means disease outbreak can cause a great loss for local farmers. Despite of high susceptibility of *P. hypophthalmus* to *Edwardsiella ictaluri*, there are not many studies on *E. ictaluri* being carried out in Malaysia. Therefore, the aims of this study are to observe the morphological appearances of *E. ictaluri* strain isolated from *P. hypophthalmus* cultured in Pahang River and to compare the biochemical characteristics of these strains with the *E. ictaluri* isolate Hawke et al. from ATCC (ATCC®33202™). Four isolates of local *E. ictaluri* were used, one isolate from wild, and three isolates obtained from caged-cultured *P. hypophthalmus* labelled as PPH 128, PTP 143, and PPH 144. Eight biochemical tests were employed i.e., are Gram-staining, motility, oxidase, catalase, sugar fermentation, pH, temperature and antibiotic sensitivity test (AST). The colonies of all isolates were small (pinpoint) and translucent on BA, BHIA and TSA. All isolates are Gram-negative with small rod-shaped cells. The isolates are motile, oxidase-negative and catalase-positive. There are various results in sugar fermentation test of each isolates while for pH test, PPH 144 grew in all three different conditions (pH 4, pH 7 and pH 10), PPH 128 and PTP 143 grew in pH 7 and pH 10 while ATCC grew in pH 7. Temperature test employed at 60°C and 120°C showed that all samples still shown properties of Gram-negative bacteria. AST showed no inhibition zone for ATCC, wild isolate, PPH 128 and PTP 143, while, PPH 144 showed inhibition zone in tetracycline (20mm), oxytetracycline (13mm) and gentamycin (10mm). Overall, the characteristics of *E. ictaluri* strain isolated from *P. hypophthalmus* cultured in Pahang River are not significantly distinct compared to *E. ictaluri* ATCC strain, thus this bacterium is considered as a homogenous species. This study will help in improving the information regarding the infection of *E. ictaluri* in *P. hypophthalmus*.

Keywords: *Pangasianodon hypophthalmus*, *Edwardsiella ictaluri*, phenotypic, characteristic

Short Biography

Dr. Rimatulhana is a Research Officer at the National Fish Health Research Division (Nafish), Fisheries Research Institute. Dr Rimatulhana Ramly completed her Masters of Aquatic Medicine from the Norwegian School of Veterinary Science, Oslo and received her PhD on Molecular Virology from the University of Life Sciences, Norway. Dr. Rimatulhana Ramly has authored several publications in various journals and books. Her publications reflect his research interests in microbiology and fish health management.

***PvRaf* knockdown using RNA interference increased survival of *Litopenaeus vannamei* infected with white spot syndrome virus**

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Shrimp aquaculture is a major source of income in the intertropical countries of the Southeast Asian region. However, a great decline in production was observed due to a major viral pathogen, the White Spot Syndrome Virus (WSSV) which continues to prevail despite many preventive measures applied to deter the virus. RNA interference (RNAi) technology has been employed to reveal functions of specific genes in the virus and its host with the aim of controlling WSSV by elucidating complex host-virus interactions. This study determined the involvement of *PvRaf*, a key component of the Ras/Raf/MAPK pathway. Raf promotes cell survival through its protein to protein interactions and MEK-ERK independent mechanism. DNA sequencing revealed that *PvRaf* has a resulting size of 1.9 kb. Phylogenetic analysis revealed that *PvRaf* is highly homologous to reported Raf genes of other organisms. Moreover, *PvRaf* is ubiquitously expressed in vital organs suggesting that it is essential to metabolic functions of the shrimp and may also play a role in its innate immune system as highlighted in its expression in the hemocytes. Quantitative Real Time-PCR (qRT-PCR) results confirmed *in vivo* gene knockdown of *PvRaf* using RNA interference. qRT-PCR showed that *PvRaf* knockdown yields downregulation of the gene in *PvRaf*-dsRNA and GFP-dsRNA treated shrimps. Gene expression analysis showed significant downregulation from day-0 to day-1 post infection in *PvRaf*-dsRNA treatment relative to the control samples. Statistical analysis of the survival data indicates that *PvRaf*-dsRNA treatment has a significant protective effect against WSSV compared to GFP-dsRNA and PBS treated shrimps.

Keywords: *Litopenaeus vannamei*, *PvRaf*, RNA interference, White Spot Syndrome Virus

***In vitro* evaluation of potential probiotics strain in biofilm formation**

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In an aquatic environment, biofilm is a combination of bacteria, microalgae, microorganisms, and extracellular polymeric substances (EPS), which function to attach and adhered to a submerged surface. Biofilm acts as a source of inorganic nutrients that can be considered a good source in promoting growth, and improving the immunity of the cultured organism. This research aims to evaluate the biofilm formation of different potential probiotic strains. Six strains; *Lysinibacillus fusiformis* strain A1 and *Bacillus* sp. strain A2 (isolated from *Amphora* sp.), *Bacillus amyloliquefaciens* strain L9 and L11 (isolated from the hemolymph of blue swimming crab), *Lysinibacillus fusiformis* strain SPS11 (isolated from *Spirulina* sp.) and *Lysinibacillus sphaericus* strain NAS32 (isolated from *Nannochloropsis* sp.) were screened for their potential in forming a biofilm. The biofilm formation assay was done by culturing the strains using a 96-well polyvinyl chloride microliter dish at 29°C without shaking. Next, the biofilm formation was sampled at 6h, 12h, 24h, 48h and 96h, stained with 1% of crystal violet and elude the dye with 90% of ethanol. Biofilm was measured at 595nm absorbance using a plate reader. Results showed that *Bacillus amyloliquefaciens* strain L9 and strain L11 had the highest biofilm formation at 24 hours with the reading of 3.56 and 3.67 respectively compared to *Lysinibacillus fusiformis* strain A1 (1.40), *Bacillus* sp. strain A2 (1.09), *Lysinibacillus fusiformis* strain SPS11(1.31) and *Lysinibacillus sphaericus* strain NAS32 (1.48) at the same time interval. Moreover, the biofilm formation of these strains showed the highest adhesive properties at 24hr and start to decrease after 24 hr. In conclusion, *Bacillus amyloliquefaciens* strain L9 and L11 showed the best formation of biofilm which is one of the main characteristics of a good probiotic.

Keywords: bacteria, potential probiotics, biofilm formation, *Bacillus amyloliquefaciens*

DAY 2: 24 August 2022 (Thursday)
Session 4: Prevention & Control Measures
(Parasitic, Bacterial & Viral Diseases)



Keynote 4

Emerging aquatic animal diseases and response actions to their emergencies in Asia and the Pacific Region

Jie Huang

Network of Aquaculture Centres in Asia-Pacific (NACA)

The presentation briefed the causative agents, affected species, and presence in the region of 14 emerging diseases and other more than 300 newly found viruses reported in finfish, crustaceans, molluscs, and amphibians in the recent decade. Strategies to facilitate the solution for emerging diseases were proposed. Challenges in the identification of causative agents for emerging diseases were discussed. A technology roadmap and approaches were proposed for the confirmation of causative agents of emerging or multiple infections. For finding emerging diseases, governments shall encourage reports from the private sector, establish a communication platform for remote diagnosis, encourage active media for the aquaculture stakeholders, and develop an aquaculture insurance policy based on reporting of aquaculture diseases. To identify emerging diseases, governance-research-services-industry cooperation is encouraged, the aquatic animal health research shall be supported, passive surveillance or rapid response to emerging diseases needs to be implemented, and it is encouraged to apply metatranscriptomic and metagenomic based technologies broadly. For emerging disease notification, publications on case studies need to be encouraged, the international mechanism trusting tradability based on transparency needs to be established, the contribution of transparency from all member sources can be accounted for, and the contribution of finding emerging diseases shall be encouraged. To better respond to emerging diseases, relevant authorities and enterprises shall establish and implement a contingency plan at different levels. Governments shall implement the original domestic site quarantine inspection policy, establish the national zoning system, and support the development and application of innovative treatment measures. More international, regional, national, and local training courses for emerging disease response need to be organized.

Short Biography

Dr. Jie Huang, a Chinese national, obtained his BSc on virology in Wuhan University in 1987, an MSc in the Wuhan Virology Institute, Chinese Academy of Science (CAS) in 1990, and his PhD on marine biology in the Ocean Institute, CAS, in 2010. He is the Director General of the Network of Aquaculture Centres in Asia-Pacific (NACA). He devotes himself to strengthening the network and its contribution to sustainable aquaculture development in the region. As an expert on aquaculture epidemiology and biosecurity, Dr. Huang previously served as a Principal Investigator in Yellow Sea Fisheries Research Institute (YSFRI), Chinese Academy of Fishery Sciences (CAFS); the Chief Scientist of CAFS on disease control; the Designated Expert of the OIE Reference Laboratories for WSD and IHHN. Dr. Huang has conducted projects on diagnostics, epidemiology, and control technology for aquatic animal diseases since 1990, reported emerging pathogens and diseases, established new diagnostic methods, developed disease prevention techniques, and actively promoted the biosecurity concept for aquaculture. He had 178 corresponding author papers, was awarded 14 patents as the first inventor, and obtained other achievements. More than a hundred doctoral and master-level students and postdoctoral scientists have completed their studies under his tutelage.

Oral Presentation

Towards heuristic design of vaccines: Transforming transposon insertion sequencing (TIS) for identification of novel in vivo virulence factors in fish pathogen

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China claims No. 1 place in the aquaculture yields in the world. However, the mariculture industries are also threatened by infectious diseases which caused huge economic losses every year. *Vibro* spp. and *Edwardsiella* bacteria represent the most threatening pathogens for farmed fish. Vaccines is a boon to protect against infection diseases. The live attenuated vaccines (LAV) stand for the most promising prevention strategy to control diseases. Recently we obtained the first two licenses in China for live attenuated vaccines against *Edwardsiella* and *Vibrio* bacterial infections in fish and widely used in the maricultures for flatfish. For development of LAV, the key is to identify suitable gene target for the construction of LAV strain. How could we avoid traditional trial and error methods but develop the rational or even heuristics designed vaccines? Here I will report the establishment of pattern analysis of conditional essential gene (PACE) driven by transposon insertion sequencing and related PACE algorithm to facilitate the success development of LAV vaccines against fish pathogens. For brief, highly saturated transposon insertion mutant library were established for *Edwardsiella* and *Vibrio*, then inoculated into fish before collect survived bacteria, and the fitness cost and abundance of each of the mutant in vivo in fish were calculated. As a proof of principle test, almost all the established in vivo virulence genes were identified by this pipeline. Moreover, many of the genes with similar pattern of in vivo fitness was proved to be of ideal LAV vaccine candidates. We reported several novel genes contributing to the in vivo infection as well we their utility in LAV vaccine development.

Keywords: PACE, *Edwardsiella piscicida*, transposon insertion sequencing, live attenuated vaccine, fish pathogen

Application of Natural Products: An Effective Approach to Control Fish Parasites in Aquaculture

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Parasitic infestation is a serious problem for different types of fish species such as groupers, hybrid groupers, snappers, seabass, etc. reared in open floating net-cages in Malaysia. Some of the commonly found parasites are monogeneans (*Benedenia* spp., *Neobenedenia* spp.), sea lice (*Caligus epidemicus*, *C. rotundigenitalis*, etc.) and leech, *Zeylanicobdella arugamensis*. Recently, the infestation of parasitic leech *Z. arugamensis* (Annelida: Hirudinea: Piscicolidae) spread rapidly in South-east Asian countries and caused economic losses. The control is vital for the management of aquaculture industry hence, chemicals such as formalin have been used. However, the best alternative is the application of natural product as a biocontrol agent since it has various metabolites with less or zero toxicity. Our recent studies aimed to elucidate the anti-parasitic efficacy of the solvent extracts of different plants from Sabah. Herbal plants found in Sabah have been selected in our experiments to kill the leeches in a natural way. The leech-infested hybrid groupers were challenged against the various concentrations of solvent extracts of the selected plants. For ex. a herbal plant, taken the average time to kill the leeches at concentrations of 25, 50 and 100 mg/ml was 25.11±3.26, 11.91±0.99, and 4.88±0.50 min., respectively. Further, at various low concentrations 2.5, 5 and 10 mg/ml, the infested hybrid groupers were disinfested in an average time of 108.33±12.65, 65.83±9.70 and 29.16±5.85 min., respectively. Those plants showed significant antiparasitic activity against *Z. arugamensis* with complete mortality. Seaweed extracts showed an excellent result against the leech. It took about 35 seconds to kill the leeches at the concentration of 100mg/ml. The biochemical composition of the extract via GC-MS and LC-Q Exactive HF orbitrap mass spectrometry indicated different antiparasitic bioactive compounds. We found some hits such as terpenoids (ivalin, isovelleral, brassinolide, and eschscholtzanthin), flavonoids (alnustin, kaempferol 7, 4'-dimethyl ether, and pachypodol), phenolics (piscidic acid, chlorogenic acid, and ankorine), and aromatics (3-hydroxycoumarin). Thus, it could be revealed that some of the Bornean herbal plants and seaweeds could be a good source of metabolites and contain vital antiparasitic phytochemical compounds to disinfest the fish for an eco-friendly aquaculture.

Key words: natural products, chemistry, plants, leech, groupers, aquaculture, agriculture, GCMS and LC-QTOF-MS, biocontrol agent

In silico selection and characterization of DNA aptamers against *Vibrio parahaemolyticus* luxP periplasmic receptor protein

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Aptamer has been studied extensively as a substitute of antibodies for various applications. It is rivalling antibody in the application of disease diagnosis and therapy. Small in molecular size and non-immunogenic properties of aptamers greatly contribute to its potential as therapeutic agent. While the aptamer binding affinity and specificity allow its application in disease detection and diagnosis. In this study, in-silico modeling was approached to design single-stranded DNA aptamers against bacterial quorum sensing receptor. The crystal structure of the apo form of *Vibrio harveyi* receptor protein (PDB ID: 1ZHH) was used as target. An initial library containing 256 short fragments of nucleotides with randomized sequences was used in molecular docking against the target protein. Aptamer candidates of eight nucleotides in length that possess significant binding affinity were selected for subsequent molecular dynamic (MD) simulation. The protein-aptamer complex was simulated for 50ns, and the complex stability was assessed. The protein-aptamer binding interaction was examined using Molecular Mechanics Poisson-Boltzmann Surface Area (MMPBSA) approach. Subsequently, selected aptamer candidates were modified by addition of complementary base to form short hairpin structured aptamers. Binding of the selected aptamers was verified by isothermal titration calorimetry (ITC), which is a frequently used technique in quantitative studies of biomolecular interactions. MD simulation revealed aptamer candidates that possess significant binding affinity to the target protein binding domain, where the calculated binding energy was lower than -250 kJ/mol. Number of hydrogen bond during 50ns MD simulation recorded a range from 5-15 bonds. Verification of protein-aptamer interactions by ITC identified two short hairpin structured aptamers to possess significant K_d value. In-vitro binding assay and subsequent preliminary analysis demonstrated anti-QS activities of the designed aptamer, which suppressed the bioluminescence of *Vibrio* that is regulated by QS cascade. Further analysis will be evaluating the suppression of virulence factor by the anti-QS DNA aptamers.

Keywords: Single-stranded DNA aptamer, Quorum sensing receptor, In-silico modelling, Quorum quenching

Potential practical application of a live attenuated vaccine against herpesviral hematopoietic necrosis of goldfish

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Herpesviral hematopoietic necrosis (HVHN) caused huge economic losses in goldfish *Carassius auratus* industry. A live attenuated vaccine was developed by propagating the causative virus, cyprinid herpesvirus-2 (CyHV-2), in ginbuna CFS cell line 7 times and further in carp KF-1 cell line 8 times (P7-P8). In this study, the vaccine administration method and optimum vaccination temperature were studied for the practical application of this vaccine. Goldfish were vaccinated with P7-P8-P1 (P7-P8 propagated once in RyuF-2 cells) by bathing at a conventional concentration (1:1000) for 2 h and subsequently challenged with virulent CyHV-2 at 21 days post-vaccination (dpv). To examine vaccine efficacy of the improved vaccine administration method, goldfish vaccinated using the showering method (1:100; 3 mL/20 fish) for 10 s were reared at 15°C, 20°C, 25°C and 30°C, which are virus permissive temperatures *in vitro*, for 3 weeks and then challenged at 28 dpv after shifting each rearing temperature to 25°C. The virus growth dynamics in organs was measured by qPCR after bath and showering vaccination. The bath vaccination showed relative percentage survival (RPS) of 84.2% and the vaccine virus growth dynamics in host after vaccination showed the virus load in the kidney and spleen peaked at 5 and 7 dpv at $10^{5.8}$ and $10^{6.3}$ virus DNA copies/mg, respectively. Vaccine virus was not detected at 21 dpv. Showering vaccinated fish reared at 15°C, 20°C, 25°C and 30°C showed 73%, 78%, 100% and 78% RPS, and peak virus load were around $10^{3.7}$, $10^{6.8}$, $10^{6.5}$ and $10^{4.6}$ virus DNA copies/mg, respectively. Taken together, the vaccine can grow in host without causing any mortality and abnormality, and consequently, was effective in fish against HVHN at the tested water temperatures. The showering vaccination method, which required small quantity of vaccine, is less laborious and cost efficient, and can be applicable in goldfish aquaculture.

Keywords: Live attenuated vaccine, Herpesviral hematopoietic necrosis, Goldfish, Vaccine administration, vaccination temperature

Short Biography

Mr. Saito is currently studying his Doctoral degree on practical application and immunological mechanism of a live attenuated vaccine against herpesviral hematopoietic necrosis of goldfish. He completed his Master's degree in Tokyo University of Marine Science and Technology, Japan and undergraduate degree in University Malaysia Sabah, Malaysia. After his study in fungal infections of mud crab in Malaysia, he extended his connections to Japan and studied viruses and vaccine applications under the supervision of Prof. Sano Motohiko. Recently, his publications in the development of this live attenuated vaccine reflect his research interests in connecting research discoveries with real world application

Active and passive immune responses and protection of Nile tilapia (*Oreochromis niloticus*) immunized with tilapia lake virus inactivated vaccines

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Tilapia tilapine virus, termed tilapia lake virus (TiLV), is a contagious viral aetiology threatening to the tilapia industry. Here, we develop two simple cell-culture heat-killed and formalin-killed vaccines (HKV and FKV) aiming to prevent this disease. The vaccine efficacies were evaluated by intraperitoneal injection on juvenile tilapia with each vaccine containing 1.8×10^6 TCID₅₀ inactivated virus. Booster doses were performed at 21-day post-primary vaccination (dppv) in the same manner. At 28 dppv, the fish were challenged with a lethal dose of TiLV. Expression of five immune genes in head kidney and spleen of experimental fish was assessed at 14 and 2-dppv and again 7-day post booster vaccination. At the same time points, TiLV-specific IgM responses were evaluated by ELISA. Then, we run the same vaccination regime on 3 treatments including four male and eight female tilapia broodstock for each with the double vaccine dose. One week after the booster, mating was performed. Broodstock blood sera, fertilized eggs and larvae were collected from 6–14 weeks post-primary vaccination for measurement of TiLV-specific antibody levels. In juvenile tilapia, the results showed that both vaccines confer significant protection for juvenile tilapia, with relative percentage survival of 71.3% and 79.6% for HKV and FKV, respectively. Significant up-regulation of IgM and IgT was observed in the head kidney of fish vaccinated with HKV at 21 dpv, while IgM, IgD and CD4 expression increased in the head kidney of fish receiving FKV at the same time point. Both vaccines induced a specific IgM response in both serum and mucus. In tilapia broodstock, TiLV-specific antibodies were generated in the majority of both male and female broodstock vaccinated with either the HKV or FKV and these antibodies can be transferred into the fertilized eggs and 1–3-day-old larvae from vaccinated broodstock, suggesting a potential strategy to prevent TiLV infection in the early stage of development.

Keywords: inactivated vaccine, tilapia lake virus, maternal passive immunity, antibody

Short Biography

Thao Thu Mai is currently a PhD candidate at the Chulalongkorn University, Thailand. She completed her Master degree in Genetics at Vietnam National University and she then worked as a researcher at Biotechnology Center of Ho Chi Minh city, Vietnam. Her research focuses on development of vaccines to prevent infectious diseases in fish. She has authored several publications in various journals.

3-Minutes Presentation

Organic acid profiles of three potential probiotics from whiteleg shrimp, *Litopenaeus vannamei*

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Various aquaculture species, particularly shrimp, suffer major economic losses due to bacterial infections. Antibiotics were utilized to solve this problem but currently prohibited due to its negative impact. Due to this, post biotic can be investigated as an alternative to antibiotics but this study is limited in aquaculture. Previous work showed that post biotic has antibacterial activities, however, its organic acid profiles has yet to be determined. Thus, the objective for this study was to determine the organic acid profiles of three potential probiotics. Three bacteria strains were selected for this study (*Enterobacter* sp. P17, *Bacillus thuringiensis* P18 and *Lactobacillus plantarum* P20). The post biotic was extracted using ethyl acetate. The organic acid content of the post biotic from each bacterium was analysed using HPLC analysis. Five organic acids (propionic acid, butyric acid, citric acid, formic acid and lactic acid) were chosen in this study. The result revealed that the butyric acid and lactic acid were present in each sample and *L. plantarum* P20 have the highest concentration of butyric acid which was 22951.3 mg/kg compared to *Enterococcus* sp. P17 (14568.5 mg/kg) and *B. thuringiensis* P18 (20799.2 mg/kg). Studies shown that organic acid such as butyric acid and lactic acid have many benefits to aquaculture as an antimicrobial and antioxidant, and can be used as an alternative to antibiotic. This study revealed that all potential probiotic have the potential to be chosen as prevention method in aquaculture and be used as supplement in aquaculture feed. As for potential candidate for the post biotic production, *L. plantarum* was a better candidate due to the high yield of organic acid produced.

Keywords: alternative to antibiotics, prevention method, *Lactobacillus plantarum*, postbiotic, aquaculture.

Short Biography

This student is currently studying as a postgraduate student at Universiti Malaysia Sabah, Kota Kinabalu. His current research title is "Efficacy of postbiotic as bacterial disease prevention for white leg shrimp (*Litopenaeus vannamei*) culture". He received his Bachelor degree of science (Hons.) biology at Universiti Teknologi Mara, Kota Kinabalu, Sabah.

Field assessments of oral *Vibrio* vaccine indicate the potential for protection against vibriosis in hybrid grouper, *Epinephelus fuscoguttatus* × *Epinephelus lanceolatus*

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Vibriosis is one of the most common threats to farmed grouper; thus, substantial efforts are underway to control the disease. This study presents an oral vaccination against vibriosis in marine fish with double booster immunization. The *Vibrio harveyi* strain VH1 vaccine candidate was selected from infected groupers *Epinephelus* sp. in a local farm and was formalin-inactivated and combined with commercial feed at a 10% ratio (v/w). A field trial was conducted on a mariculture farm in Langkawi Island, Malaysia, to study the effect of field vaccination using the oral *V. harveyi* strain VH1 vaccine candidate. A total of 3000 hybrid grouper juveniles were divided into two groups in triplicate. Fish of Group 1 were not vaccinated, while Group 2 were vaccinated with the feed-based vaccine. Vaccinations were carried out on days 0, 14, and 42 via feeding the fish with the vaccine at 4% body weight for five consecutive days. Subsequent administration of the first and second booster for five consecutive days, starting on days 14 and 42, improved the specific antibody level in serum, mucus, and gut lavage fluid and reached a highly significant ($P < 0.05$) value on day 56 before slightly decreasing onwards. Antibody titers of the control unvaccinated group remained relatively stable and low throughout the experimental period. At the end of the study period, the fish survival rate was 80% for the vaccinated group, significantly ($P < 0.05$) higher than the 65% seen in the control unvaccinated group. Furthermore, the vaccinated fish showed better growth performances ($P < 0.05$). Therefore, the oral *Vibrio* vaccine from the inactivated *V. harveyi* strain VH1 is a potential vaccine candidate that could stimulate good immune responses and confer high protection in farm hybrid grouper *Epinephelus fuscoguttatus* × *Epinephelus lanceolatus*.

Keywords: Vibriosis, *Epinephelus fuscoguttatus* × *Epinephelus lanceolatus*, oral vaccine, marine fish

Short Biography

Aslah Mohamad is currently a PhD student at Universiti Putra Malaysia (UPM). He completed his Master's degree in Aquaculture from Universiti Malaysia Terengganu (UMT) in 2017. He is currently an enumerator at UPM, working on several fish vaccines. Aslah Mohamad has authored several publications in various journals and proceedings. His publications reflect his research interests in fish diseases and vaccinology.

Field efficacy of a feed-based Inactivated *Vibrio harveyi* vaccine against vibriosis in cage-cultured Asian seabass, *Lates Calcarifer*

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Vibrio harveyi is an important aquaculture pathogen that causes vibriosis and infects a large number of fish species. Vaccination is one of the alternatives to overcome disease outbreaks and was reported to be more effective and safer for humans and the environment than antibiotics. In a previous experiment, this vaccine showed promising results where the vaccinated fish was found to have an RPS of 70% to 85% against multiple *Vibrio* spp. such as *V. harveyi*, *V. alginolyticus* and *V. parahaemolyticus*. In this study, the field trial was conducted at Pulau Ketam, Klang, Selangor, where this newly developed feed-based killed *V. harveyi* vaccine was used to study the immune response and survival of this kind of vaccine in Asian seabass. A total of 4800 Asian seabass, *Lates calcarifer* of 182 ± 31 g, were separated into two cage groups in duplicate. Fish of Group 1 were not vaccinated, and Group 2 was vaccinated with feed-based killed *Vibrio harveyi* vaccine. Vaccinations were done on weeks 0, 2 and 6 orally using the feed-based bacterin vaccine at 4% body weight. Samples of serum, mucus and gut lavage fluid for antibody study were collected at 14-day intervals throughout the 16-week study period. Following vaccination with the feed-based vaccine, IgM antibody levels showed a significant ($p < 0.05$) increase in the vaccinated group as early as week 2 and peaked at week 8 before slightly decreasing afterwards. At the end of the study period, the growth performance showed a significant ($p < 0.05$) increase in the vaccinated group with lower bacterial isolation in the gut than the unvaccinated group. Nevertheless, the vaccinated fish survival rate was 71.3% higher than the 67.7% in the unvaccinated fish. In our opinion, this feed-based vaccine could potentially stimulate good immune responses and provides good protection in cage cultured Asian seabass, *Lates calcarifer*.

Keywords: Vibriosis, *Lates calcarifer*, feed-based vaccine, marine fish

Short Biography

Zahaludin Amir-Danial is currently an MSc student at Universiti Putra Malaysia (UPM). He completed his Bachelor's degree in Applied Science (Fisheries) from Universiti Malaysia Terengganu (UMT) in 2019. Currently, he is a special graduate research assistant at UPM, working on several fish vaccines. Zahaludin Amir-Danial has authored several publications in various journals and proceedings, and his publications reflect his research interests in fish diseases and vaccinology.

***In vitro* antimicrobial activity of plant extracts against fish pathogenic bacteria**

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Antibiotic resistance has emerged as a result of the advent of new diseases and the rising usage of antibiotics in aquaculture. Hence, the aquaculture industry is paying more attention in finding new antibacterial compounds from natural sources to tackle this problem. Antibacterial multidrug resistance in aquaculture systems necessitates the investigation of alternate therapies for the treatment of diseases induced by these drugs. Plant extracts have been recommended as one of these alternatives; they have showed antibacterial action in studies *in vitro* and *in vivo* in fish and shellfish. This study reports *in vitro* antimicrobial activity of selected plant extracts against fish pathogenic bacteria in aquaculture. The research method employed in this study is an experimental research approach with a completely randomised design with extract concentrations of 0%, 20%, 40%, 60%, 80%, and 100%. The antibacterial activity and minimum inhibitory concentrations (MIC) of plant extracts against pathogenic bacteria were determined using the agar well diffusion method. Plant extracts showed antibacterial activity against some of the bacteria, and results indicate an MIC and MBC showed inhibitory activity occurred against pathogenic bacteria. This study hence revealed the potential of plant extract to be used as an alternative treatment in aquaculture.

Keywords: Antimicrobial activity, plant extracts, pathogenic bacteria, alternative treatment.

Short Biography

Scientist is currently working as research officer at Fisheries Research Institute Bintawa, Kuching Sarawak Malaysia. She received her Bachelor degree on Agrotechnology Aquaculture from the Universiti Malaysia Terengganu. She has authored several publications and her publications reflect her research interests in fish disease.

Control of *Aeromonas* sp. infection in nursery stage of *Pangasius nasutus* (Bleeker, 1863) through bio-encapsulation of herbal extract; SirehMAX in live feed

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A study was carried out to develop *Pangasius nasutus* broodstock with high survival trait following previous mortalities caused by bacterial infection in nursery stage. Study was done by stocking 500 *P. nasutus* fingerlings in 100 L tanks and were fed twice daily with live feed, *Moina* sp. Two doses of SirehMAX; an ethanolic extract of betel leaves were encapsulated in *Moina* sp. which were 100 and 150 mg/L while 0 mg/L as negative control. Study was done in triplicates during a period of four weeks. After one week, *Aeromonas* prevalence for treatment tanks (*Moina* + SirehMAX) 100 (T) and 150 mg/L (S) were at $22.22 \pm 25.46\%$ and $66.67 \pm 34.7\%$; respectively compared to $38.89 \pm 34.7\%$ in negative control tanks (*Moina* only). *Aeromonas* prevalence for T and S tanks decreased on the second week to $0.00 \pm 0.0\%$ and $11.11 \pm 19.24\%$; respectively compared to $5.56 \pm 9.62\%$ for negative control. On the third week, *Aeromonas* prevalence for T tanks slightly increased to $11.11 \pm 9.62\%$ while S tanks remained at $11.11 \pm 19.24\%$ compared to negative control which also remained unchanged. *Aeromonas* prevalence on the final week; however, decreased to $5.56 \pm 9.62\%$ and $0.00 \pm 0.0\%$ for T and S tanks compared to $5.56 \pm 9.62\%$ for negative control tanks. Comparatively, there are no significant difference between bacterial control regime between 100 and 150 mg/L SirehMAX, however; mortality rate for 150 mg/L tanks were $15.00 \pm 11.39\%$; lower than $17.00 \pm 12.65\%$ for 100 mg/L and $21.87 \pm 15.65\%$ for negative control tanks. This study showed that SirehMAX encapsulated in live feed might be able to control the *Aeromonas* infection in *P. nasutus* nursery culture and increasing fingerlings survival.

Keywords: SirehMax: *Pangasius nasutus*: encapsulated: live feed: *Aeromonas*

Short Biography

Mr. Iftikhar Ahmad bin Abdul Rafi is currently working as a Research Officer at the Freshwater Fisheries Research Division, Department of Fisheries Malaysia. He completed his Masters in Health Science (Pharmacy) from the National University of Malaysia. He then worked at the Crustacean Aquaculture Research Division Pulau Sayak and National Health Research Centre Division before serving as Research Officer at the Freshwater Fisheries Research Division Glami Lemi, Jelebu, Negeri Sembilan. Mr Iftikhar Ahmad has authored several publications in various journals and books. His publications reflect his research interests in aquatic animal health.

Assessment of inhibitory properties of potential probiotics isolated from *Pangasius nasutus* against *Aeromonas hydrophila* and *Streptococcus agalactiae*

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Aquaculture activity is in need of a more environmentally friendly approach to enhance the immune system, promote growth and survival in various cultured aquatic organisms including *Pangasius* species. The survival of the *Pangasius* species has been reported to be affected by bacterial diseases. Therefore, this study aims to evaluate potential probiotics isolated from *Pangasius nasutus* against *Aeromonas hydrophila* and *Streptococcus agalactiae* as a safer alternative to antibiotics. Six healthy *P. nasutus* were dissected to isolate potential probiotics from the internal organs (intestine and stomach). A total of 70 bacterial strains were successfully isolated from the organs (21 strains from the stomach and 49 strains from the intestine) and subjected to further screening. *In vitro* antagonism assays which include spot and well diffusion were done using the isolated strains against pathogens (*A. hydrophila* and *S. agalactiae*). Through preliminary screening, five strains demonstrated the ability to inhibit the growth of pathogen *A. hydrophila*. Strain S1 and L1 inhibited the growth of *A. hydrophila* with the inhibitory zone of 1.5±1mm and 1.2±1mm respectively. Meanwhile, strain L2, L8 and L12 inhibited the growth of *A. hydrophila* with the inhibitory zone of 1.0±1mm, 1.2±1mm and 2.0±1mm respectively. Out of the five potential probiotics, only strain L2 was able to inhibit the growth of *S. agalactiae* with an inhibitory zone size of 1.0±1mm. These five strains were identified using 16s rRNA gene sequencing. The strain S1, L1, L2, L8 and L12 were identified as *Lactococcus lactis*, *Weissella confusa*, *Cosenzaea myxofaciens*, *Lactococcus garvieae* and *Plesiomonas shigelloides* respectively. The potential of these strains could be further studied to understand their mechanisms and full potential as probiotics for *P. nasutus* culture.

Keywords: *Pangasius nasutus*, potential probiotic, *Aeromonas hydrophila*, *Streptococcus agalactiae*, inhibitory properties

Ultrafiltration membrane technology for virus mitigation control strategy in indoor shrimp farming system

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The shrimp farming industry globally faces numerous challenges, most importantly various viral, bacterial and fungal diseases. One of the major viruses in shrimp industry is White Spot Syndrome Virus (WSSV) which can cause up to 100% mortality. As wastewater recirculation is frequent, virus removal in aquaculture wastewater is vital. In this context, an effective water recirculating system is vital to ensure the circulated water is free of bacteria, parasites and viruses. Therefore this current study intended to assess the viability of ultrafiltration (UF) membrane system for removing viruses from shrimp farming water. Molecular weight cut-off (MWCO) poly-vinylidene fluoride (PVDF) membranes of 20 kDa, 40 kDa, and 60 kDa were utilised to determine the optimal membrane pore size for WSSV removal. Polyethylene glycol (PEG) was utilised to simulate the same-sized virus in shrimp farming effluent. Consequently, it was determined that 30 kDa is a sufficient membrane MWCO for the filtering process under-regulated operating settings of 3 L/min cross flow rate and 0.3 bar trans membrane pressure. The permeate generated was analysed for pH, turbidity, total dissolved solids (TDS), biochemical oxygen demand (BOD), and dissolved oxygen (DO) to assess the water quality; the results indicated that the filtration procedure greatly improved the water quality. Disinfection can be used as a coupling approach to provide a second barrier for further ensuring stringent water quality control. A feasible membrane filtration-Disinfection system with suitable membrane material to mitigate biofouling designed for total WSSV removal. Thus, the shrimp farming industry will evolve into a sustainable and green system, reducing losses in production due to viral diseases.

DAY 3: 25 August 2022 (Thursday)
Session 5: Trends in Fish Health Management



Keynote 5

Malaysian experiences: Environment-friendly alternatives to chemicals in aquatic animal health management

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Aquaculture expansion is typically correlated with culture intensification, resulting in overcrowding and poor water quality, which promotes pathogen propagation and increase in disease outbreaks as well as mortality. Veterinary chemicals are used as prophylaxis and therapeutic of diseases in order to avoid economic losses. However, most chemotherapy causes antimicrobial resistance (AMR), and could affect the efficacy of treatment of diseases in aquatic animals health. In Malaysia, R&D on alternative medicine particularly plant extracts and commercial oils has been conducted and evaluated since the year 2010. Exploratory studies on the potentially environment-friendly alternatives to chemicals include manipulation of science-based know how in fish disease development, pathogen life cycle, and development of fish diets. Based on these concepts, Break & Protect (BP) which is a device for trapping and removing marine leeches from infested farmed fish by disrupting its life cycle was developed. Later, an improvised version (BP2) was introduced for additional 14 species of farmed fish. Another product was developed in 2017 known as KRIPeK which is a portable kit for treating fish swim bladder disorder on site. Besides that, there were also development of several plant based antimicrobial solutions such as SirehMAXTM, GARLEX, and SitroProTM for preventing and treating bacterial diseases in fish beside stimulating and promoting weight gain when administered to cultured fish. Application of commercial essential oils such as cinnamon oil was also demonstrated to be beneficial in preventing and treating marine ectoparasites. The environment-friendly alternatives presented in this paper have good potential to be applied in aquaculture for alternatives treatment and intervention of disease outbreaks.

Keywords: Local plant, commercially herbs, prevention, treatment, alternative

Short Biography

Dr. Kua Beng Chu is a senior researcher and currently working as a senior director of research at the Fisheries Research Institute, Department of Fisheries Malaysia. She received her Doctoral degree in Fish Parasitology at the age of 32 years old from the University of Science, Malaysia. Since 1996, she has been in charge of research and development in fish health with a focus on aspects of fish parasites, pathology, disease management. She has written more than 100 technical papers in journal writing, book, and successfully patented four findings.

Oral Presentation

IgY as immunoprophylaxis in aquaculture

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Due to the lack of an adaptive immune system in invertebrates, immunoprophylactic approaches like vaccination are not plausible in preventing disease outbreaks in prawn and shrimp cultures. Passive immunity is the process of providing pre-formed antibodies to confer immediate protection against infections. Hen yolk antibodies (IgY) are a promising candidate for passive immunisation adopted in the human and veterinary industry to replace the use of antibiotics. To explore the potential of passive immunity in terms in aquaculture, the present study immunised two healthy hens with LuxP protein, an identified quorum sensing (QS) molecule of *Vibrio* spp. The IgY antibodies were purified from the hyper immunised hens. The quorum quenching properties of the immune IgY were further tested with QS inhibition assays. The quorum sensing *Vibrio harveyi* BB152 strain was cultured alone as negative control while other bacterial cultures were co-cultured with various concentrations of antibody IgY (10%-60%) to determine the luminescence activity. The luminescence activities decreased from 0% IgY (control) to 60% IgY co-culture for immunised IgY, where the EC50 was at 30% IgY, indicating that purified IgY exhibited quenching activity. Two types of oral IgY immunoprophylaxis have been made, namely incorporation of IgY into feeds with the optimised binder; and bioencapsulation of IgY with *Moina*. Both *in vitro* and feeding trials showed high immunoprotective activities of IgY upon gastrointestinal digestion. The oral administration of IgY designed in the present study possesses the potential to be used as immunoprophylaxis for aquaculture.

Keywords: Passive immunisation, IgY, Quorum sensing, Quorum quencher, Bioencapsulation,

Short Biography

Dr Chong Chou Min is currently working as a senior lecturer at the Department of Aquaculture, Faculty of Agriculture, Universiti Putra Malaysia. He received his Doctoral degree or PhD in immunology from Universiti Putra Malaysia. Dr Chong has authored several publications in various journals and books. His publications reflect his research interests in fish immunology and aquaculture health management.

The effects of oral feed-based vaccination against vibriosis on gut microbiome of Asian seabass

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Vibriosis is a disease that affects Asian seabass and other farmed marine fishes, causing either chronic low-grade fatalities or severe outbreaks that result in mass mortality. Vaccination is among the most effective ways to combat disease in fish. This study describes the effects of oral feed-based vaccination against vibriosis on gut microbiome of Asian seabass. Asian seabass that cultured in floating cages were divided into two groups, which is non-vaccinated group that fed with commercial pellet and vaccinated group that was fed with commercial pellet which incorporated with the vaccine. Asian seabass was cultured through 16 weeks of vaccine regimes where the vaccine was given at week 0, week 2 and week 6. Gastrointestinal content was collected on week 0, week 2, week 6 and week 10, then proceed with DNA extraction and high-throughput sequencing. The results revealed the alpha diversity of both non-vaccinated and vaccinated group showed significant difference ($p < 0.05$) in diversity and richness of microbiota and further supported by PCoA with ANOSIM R value of 0.288. Phylum Proteobacteria was the most dominant in non-vaccinated group, followed by Firmicutes and Fusobacteria, while in vaccinated group, Firmicutes was the most dominant and followed by Proteobacteria and Fusobacteria. At genus level, non-vaccinated group dominated by *Vibrio*, while vaccinated group were dominated by *Clostridium*. Further identification of *Vibrio* to species level showed that *Vibrio harveyi* was dominantly present in non-vaccinated group compared to the vaccinated group. Lefse analysis identified 15 potential taxa markers that differentiated non-vaccinated and vaccinated Asian seabass, where LDA score of seven taxa was associated with non-vaccinated group and eight of the taxa was associated with the vaccinated group. This study revealed that gut microbiome of the Asian seabass showed different pattern of bacteria domination for each non-vaccinated and vaccinated group, indicating potential application of feed-based vaccination strategy in fish culture industry.

Keywords: Asian seabass, 16S amplicon, feed-based vaccine, vibriosis, disease

Short Biography

Jumria Sutra is currently doing her PhD at Department of Biology, Faculty of Science, Universiti Putra Malaysia. Jumria completed her Master of Science in Aquatic Biotechnology from the Universiti Putra Malaysia. She has authored several publications in various journals. Her publications reflect his research interests in aquatic microbiology and omics study.

Ectoparasites recovered from whole cage freshwater treatment on cultured marine fish disease outbreak in floating cages

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Multi-species marine finfish are farmed in floating cages, with farmers experience increasing frequent fish disease outbreaks, especially during the first year of culture. Common symptoms observed are scratches on body surface, scale drop, fin-and tail-rot, head region devoid of scales, eye opaque and blind. Ectoparasites on body surface on cultured marine fish had been well documented by Leong (2014). Farmers freshwater treat the whole cage of fish when disease outbreak occurs. Sample of ectoparasites in the container were collected for identification and counting under dissecting microscope. Three groups of ectoparasites recovered in these monitoring studies are capsalid monogeneans, (especially, *Benedenia lutjani*, *Benedenia epinepheli*, *Benedenia* spp. *Neobenedenia melleni*, *Neobenedenia* spp.), leech, (*Zeylanicobdella arugamensis*) and caligid copepods (especially, *Caligus rotundigenitalis*, *Caligus* spp.). The capsalid monogeneans, especially *B. lutjani*, are the most numerical dominant ectoparasites removed from whole cage of snappers and leech, in groupers. Regular freshwater treatment prevents frequent disease outbreak. Observations on initial disease outbreak show injuries on body surface caused by activities of large numbers of ectoparasites. These ectoparasites are considered as PRIMARY PATHOGENS initiating disease outbreaks in floating cages. The affected fish rub the body surface against the net due to irritation to rid these primary ectoparasites. Physiological complications and secondary bacterial infection result in fish dying. Crimson snappers are the numerical dominant fish species culture and the major contributor of capsalid monogeneans for infection to other fish culture. Groupers are the major contributor of leeches for infecting to other fish. This constant removal and re-infection of the ectoparasites pathogens in the cultured fish appear to be a process of natural life vaccination from the infection of these ectoparasites acting as vaccine. The affected fish become immune to these infections resulting in fewer disease outbreaks as the fish grow.

Keywords: Primary pathogens, *Benedenia lutjani*, *Benedenia epinepheli*, *Neobenedenia melleni*, *Zeylanicobdella arugamensis*, *Caligus rotundigenitalis*.

Short Biography

Dr. Leong received his Ph.D. from the University of Alberta, Canada and was a Professor of Parasitology at Universiti Sains Malaysia. His major research interests are on parasites, diseases and health management in marine fish culture in floating cages. He began these researches in 1986.

The prospect of using aptamers and digital aptasensors for real-time surveillance of diseases in aquaculture

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Aptamers are a category of oligonucleotides that is artificially synthesized and able to bind selectively to a wide variety of molecular targets. Consisting of either RNA or DNA sequences, the oligonucleotides will fold naturally into three dimensional structures that can conform to the energies and shapes of the target molecules. Another plus point is that aptamers can be easily adapted to existing diagnostic methods such as ELISA, Western Blot, biosensor etc. Aptamers can be designed either via wet lab method known as SELEX (Sequential Evolution of Ligands by Exponential Enrichment) or via our biocomputational APTCAD method. Both methods have their strengths and weaknesses; therefore, are seen to be complementary of each other. The potential of aptamer-based biosensor or APTASENSOR for real-time surveillance of animal diseases in the field will be explored in this presentation. We envision an eco-system where aptamers can be digitally designed, synthesized in-situ using a molecular printer, validated in wet lab and incorporated into portable APTASENSOR devices for field use.

Keywords: Aptamer, aptasensor, biosensor, diagnostics, disease

Short Biography

Mr. Tang Kok Mun is the co-founder and CEO of Biogenes Technologies, a Malaysian-based company focusing on development and commercialisation of molecular diagnostic technologies for applications in human healthcare, animal healthcare, agriculture, aquaculture, food safety and pollution monitoring. Mr. Tang have over 15+ years of experience in bringing technologies from R&D stage to commercial markets. Mr. Tang majors in Chemical Engineering and MBA from University of Malaya, as well as having graduated from Stanford University Center of Professional Development in Entrepreneurship. He currently serves as Industrial Fellow in Universiti Sains Malaysia Institute for Research in Molecular Medicine (INFORMM).

Zoonotic potential of Group B *Streptococcus* in aquaculture

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Streptococcus agalactiae or Group B *Streptococcus* (GBS) has been shown in the past to be a major cause of sepsis in neonates and older adults with co-morbidities. In Southeast Asia, GBS is also a threat to aquaculture and food security, causing diseases and high mortality in farmed fish, especially tilapia (*Oreochromis* sp.). GBS serotype III sequence type (ST)283 is the only strain that is known to cause food borne illness in humans. In 2015, Singapore faced a major outbreak of GBS caused by fish borne GBS ST283, yet major data gaps exist. It is still unclear how GBS ST283 spreads between fish and humans. Recently, FAO in 2021 recognized GBS ST283 as a pathogen of concern and as a threat to aquaculture in Southeast Asia. Reports of healthy asymptomatic GBS ST283 carrying fishes do exist, which suggest a potential threat as some species of food fish may serve as reservoir hosts for GBS ST283. Understanding on the transmission of GBS ST283 in food fishes and humans may prove vital in connecting links that will explain how the zoonotic spill-over occurs. Human diseases caused by fish-borne pathogens are not well documented and are no longer considered speculative. Public awareness on the importance of GBS zoonoses, their connection to aquaculture and human health is fairly low. This review focuses on evidence from literature concerning GBS ST283 infections in fishes and humans. The purpose of this review was to foster greater awareness on the zoonotic potential of GBS in aquaculture.

Keywords: Group B *Streptococcus*, ST283, transboundary, zoonotic spill-over, aquaculture

Short Biography

Mr. Snehashish is currently a PhD candidate at Universiti Putra Malaysia. He received his Master's Degree in Zoology from the Banaras Hindu University, India. He is currently working on 'Pathology and pathogenesis of Group B *Streptococcus* serotype III ST283 in tilapia (*Oreochromis* sp.) and human cell line models' in Universiti Putra Malaysia.

Refolded recombinant major capsid protein (MCP) from Infectious Spleen and Kidney Necrosis Virus (ISKNV) effectively stimulates serum specific antibody and immune related genes response in Nile tilapia (*Oreochromis niloticus*)

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Infectious spleen and kidney necrosis virus (ISKNV) is a causative agent of high mortality in fish resulting in significant economic loss to the fish industry in many countries. The major capsid protein (MCP) (ORF006) is an important structural component that mediates virus entry into the host cell, therefore it is a good candidate antigen of ISKNV for subunit vaccine development. In this study, MCP of ISKNV was successfully produced in *Escherichia coli* strain Ril and was purified as the soluble form by refolding recombinant MCP using urea in combination with dialysis process. The refolded recombinant MCP protein had ability of oligomerization to become trimer like native MCP protein. Fish immunized with refolded recombinant MCP showed significantly higher serum antibody titer than fish immunized with insoluble form of the protein ($p < 0.05$) at 21, 28- and 35- day post-immunization (dpi). Analysis of immune-related genes response in spleen and kidney of fish immunized with refolded recombinant MCP suggested that MHC-I, MHC-II, IL-1 β and IL-4 genes were also significantly expressed relative to the group immunized with insoluble protein ($p < 0.05$) at 14, 21, 28- and 35-day post immunization. The highest serum antibody and immune related genes response were found at 28 day post immunization. Therefore, refolded recombinant MCP should be better than previously reported insoluble form as the candidate subunit vaccine to prevent infection of Nile tilapia from ISKNV.

Phytobiotic-based additive for consistently improved gut integrity and health across species

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A healthy gut, both structurally (gut integrity) and functionally (gut function), is extremely important in order to ensure proper growth and health. The gut processes antinutritional factors that induce inflammation and is also the first entry point for infectious pathogens. Reinforcing gut protection is a key strategy to maintain fish immunocompetence, minimize the severity of infections, and maximize feed utilization. Gut integrity and function were evaluated following the dietary supplementation of a phytobiotic-based additive to two fish species of global importance: Gilthead seabream (*Sparus aurata*) and Nile tilapia (*Oreochromis niloticus*). Parameters included in the study were: (1) electrophysiology; (2) histopathology; and (3) proteomics. The study presents a comprehensive interpretation of the results for these key parameters, showing a consistent positive effect of the phytobiotic-based additive on improving and maintaining gut integrity and health across species. These results shed light on the efficacy of the additive in field conditions when applied during critical periods of the production cycle.

Keywords: gut health, gut integrity, marine fish, tilapia, electrophysiology

Short Biography

Dr. Ruth Garcia Gomez is currently working as Business Development Manager for health and farm care products in the Asia-Pacific region under the Adisseo Aqua Team. She holds a BSc in veterinary science and a PhD in veterinary science-aquatic health management. She has been working as aquatic health and aquatic biosecurity specialist for 20 years with various companies, research institutions and international organizations. She has a strong experience in the design and implementation of integrated health management approaches and aquatic biosecurity strategies towards the minimization of disease pressure in aquaculture farming.

Effects of *Piper betle* extract supplementation diet as a natural antibiotic growth promoter (NAGP) to kelah (*Tor tambroides*)

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The present study was conducted to evaluate the potential of *Piper betle* extract as an herbal product as a feed additive and a natural antibiotic growth promoter (NAGP) toward kelah (*Tor* sp.) on antimicrobial activity and growth performance. In this study, for antimicrobial activity, several concentration levels of *Piper betle* extract were tested to observe the optimum dosage for bacterial inactivation of *Aeromonas hydrophila* obtained from the kelah specimen. Then, in the growth study, two treatment methods were used, which were once a month (S1) and fortnightly (S2) feedings of a diet treated with 100 ppm *Piper betle* extract, with a non-treated *Piper betle* extract diet serving as a control (C). All treatments were conducted in duplicate and the fish were stocked at 10 fish m⁻² in 3MT tanks. Fishes were fed ad libitum, twice daily. The results showed the optimal dosage for *Piper betle* extract was identified at 100 ppm with a significant difference ($P > 0.05$) in the size of the *Aeromonas hydrophila* bacterial inhibitory zone, with diameters of 18.7 ± 0.6 mm. While at 182 days of growth study, the results show that the best growth performance of kelah according to body weight gain (BWG) was treatment S2 with 131.9 ± 9.1 %, followed by S1 and control at 104.8 ± 10.2 % and 84.2 ± 9.5 %, with a significant difference ($P < 0.05$). While the S2 treatment significantly showed the best feed conversion ratio (FCR) compared to the S1 and control treatments, with 3.57 ± 0.17 , 4.27 ± 1.13 , and 5.02 ± 0.49 , respectively. In overall, there was no significant difference in survival rate among all treatments in this study. Thus, the results suggest that *Piper betle* can be considered as a NAGP in the diet at the optimum dose of 100 ppm, with fortnightly application as the best feeding regime for kelah culture.

Keywords: *Piper betle*, kelah (*Tor* sp.), natural antibiotic growth promoter (NAGP), antibacterial, growth performance

***In-vitro* responses of peritoneal macrophages of marine red hybrid tilapia (*Oreochromis* spp.) model to vibriosis: A comparative study between vaccinated and non-vaccinated fish**

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Vibriosis, an infection caused by Gram-negative bacteria of the genus *Vibrio* is a major disease in global aquaculture. Vaccination is an effective preventive measure against vibriosis. Phagocytosis, the fundamental cellular mechanism that eliminates invading pathogens is governed by phagocytic cells like macrophages and neutrophils. To gain insight into the mechanism of vaccination and its role in influencing macrophage activity in reducing the number of live *Vibrio* species prior to the establishment of infection in the host, *in vitro* responses of peritoneal macrophages of vaccinated and unvaccinated marine red hybrid tilapia were compared in terms of phagocytosis, intracellular killing, and macrophage death rates. A total of 90 healthy marine red hybrid tilapia were divided into three equal groups. Group 1 and 2 were vaccinated using feed-based oil-adjuvanted and non-adjuvanted killed whole cell *V. harveyi* vaccine, respectively. Group 3 was not vaccinated. Vaccine feed was given on weeks 0, 2 and 6. Peritoneal macrophages were collected at week 0 and 10. Macrophages were exposed to *V. harveyi* or *V. alginolyticus* and harvested at 0-, 30-, 60- and 120-minutes post-infection. The rate of phagocytosis, intracellular killing rate of bacteria and rate of macrophage cell death were calculated. In general, post-vaccinated macrophages of all treatment groups showed a significant ($p < 0.05$) increase in phagocytosis than pre-vaccinated macrophages following *V. harveyi* and *V. alginolyticus* challenge. Macrophages of post-vaccine oil-adjuvanted group showed significant ($p < 0.05$) increase in intracellular killing of *V. harveyi* and *V. alginolyticus*. Macrophages of post-vaccinated Group 2 fish showed higher cell death rate following *V. harveyi* and *V. alginolyticus* infection than that of post-vaccinated Group 1 fish. This study proved that the macrophages of tilapia immunized with oil-adjuvanted vaccine were more efficient in phagocytosis, intracellular killing, and more resistance compared to non-adjuvanted vaccinated and unvaccinated tilapia in the presence of *V. harveyi* or *V. alginolyticus*.

Keywords: *Vibrio harveyi*, *Vibrio alginolyticus*, vibriosis, marine tilapia, fish peritoneal macrophages, immunofluorescence

Short Biography

Dr. Manchanayake is a PhD student at the Faculty of Veterinary Medicine, University Putra Malaysia. She completed her MSc in Aquatic Resource Management and Aquaculture from the University of Peradeniya, Sri Lanka. She served as a Veterinary Researcher at the Veterinary Research Institute, Sri Lanka. Dr. Manchanayake has authored several publications in various journals and books. His publications reflect his research interests in Veterinary Pathology.

3 Minute-Presentation

The effect of different concentration of probiotics (lacto-sacc) mixtures on growth performance and feed utilization of empurau (*Tor tambroides*) fingerlings

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Tor tambroides, popularly known as Empurau, is one of the most valuable freshwater fish in Malaysia and indigenous to Sarawak. Empurau has a high level of aquaculture production due to its economic and conservation potential and high market demand. However, empurau is a slow-growing fish, result in high feeding cost for this fish culture. This study evaluated the effect of different concentrations of probiotics (Lacto-sacc) mixtures on growth performance and feed utilization of Empurau (*Tor tambroides*) fingerling. Lacto-sacc is a probiotic combination of *Lactobacillus acidophilus* and *Saccharomyces cerevisiae* that is frequently used as a feed additive and antibiotic replacement in livestock diets. Fingerlings stocked at 50 fingerlings/tank with average weight of 6.5g. Diets were formulated with 40% protein and supplemented with different concentration of Lacto-sacc forming 4 experimental diet, control diet (T_A), 0.5% Lacto-sacc (T_B), 1.0% Lacto-sacc (T_C) and 1.25% Lacto-sacc (T_D). The fingerlings were fed twice a day with 5% of the average body weight for 20 weeks. As a result, no significant difference was found between treatments. However, empurau fed with T_B diet has the best results for almost all parameters studied. In comparison to other treatments, T_B exhibits the greatest weight gain with an increment of 11.66g, the highest specific growth rate (SGR), and the lowest feed conversion rate (FCR). The control (T_A) exhibited the lowest survival rate (SR) and SGR, with the highest FCR. According to observation, mortality in T_B and T_D were overfeeding of larger fish. In contrast, control group's (T_A) mortality is caused by disease infection with pop-eye and red spot on the skin. The study shows fingerlings supplied with Lacto-sacc, especially at 0.5%, show better growth performance and feed utilization than the control diet that not supplemented with Lacto-sacc.

Keywords: Empurau, *Lactobacillus acidophilus*, Lacto-sacc, *Saccharomyces cerevisiae*, *Tor tambroides*

Short Biography

Ms. Sing Ying Chua is currently a Master student in Aquatic Science at Universiti Malaysia Sarawak, majoring in field of aquaculture. She finds her interest in aquaculture field during internship in Chao Jing Aquaculture centre, National Museum of Marine Science and Technology. During her degree final year project, she includes in aquaponics project. A journal about that aquaponics project was published.

The effect of commercial cinnamon essential oil (EOCIN) against bacterial infection in farmed red snapper (*Lutjanus argentimaculatus*)

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Bacterial infections known to causes high mortalities and great losses to the farmers. The threats of bacterial infection in aquaculture raise the interest in finding new treatments. There is a new trend of using essential oils as an antibacterial. Thus, the objective of this study is to evaluate the effect of cinnamon essential oil (EOCIN) towards bacterial infection in farmed red snapper (*Lutjanus argentimaculatus*). The study was carried out for 43 days, started on 23 November 2021 in Jerejak Island cage culture. Fish with average size of 18.64 ± 3.31 g were divided into 2 groups; control and treatment. Control fishes were fed with normal pellets while treatment fishes were fed with pellets mixed with EOCIN for 14 days consecutively at 1.5% (v/w). After completion of treatment, fishes were sampled on day 15, 29, and 43. A total of 20 control fishes and 20 treatment fishes were sampled each sampling session. The fish were dissected and the samples from internal organ; spleen, liver and kidney and were cultured on tryptic soy agar (TSA) with additional 5% salt. Gram staining was conducted and the identification of bacteria was performed using API method (Biomeriux). The results showed lower prevalence percentage of bacteria found in the internal organs of fish of EOCIN treated group compared to the control (Day 15); *Vibrio vulnificus* (5% treatment vs. 25% control), and *Vibrio parahaemolyticus* (5% treatment vs. 10% control). However, on Day 29 and Day 43 (after completion of EOCIN treatment), increasing pattern of bacterial prevalence can be seen in the EOCIN group compared to the control; *Vibrio vulnificus* (5% treatment vs. 6% control), *Vibrio parahaemolyticus* (5% treatment vs. 6% control) and *Photobacterium damsela* (50% treatment vs. 39% control). This preliminary study showed that administration of EOCIN to farmed red snapper reduced the prevalence of bacterial found in the internal organs of fish, thus reduce the exposure and risk of bacterial infection to cultured fish. However, prolong effect of EOCIN cannot be seen. Thus, further study needs to be done to confirm the best concentration and period for EOCIN administration.

Keywords: cinnamon essential oil (EOCIN), red snapper, bacterial prevalence

Short Biography

Ms. Noor Hanis Abu Halim is currently working as Research Officer at National Fish Health Research Centre, Batu Maung, Penang, Malaysia. She completed her Master Degree of Science in Biotechnology from Universiti Malaysia Sabah (UMS). She then worked at Universiti Sains Malaysia (USM), served as Research Assistant at Regenerative Medicine Cluster, Advanced Medical and Dental Institute (AMDI), USM, Penang. She currently pursuing her PhD in Cell and Molecular Biology. She has authored several publications in various journals and books. Her publications reflect her research interests in Biotechnology and Fisheries.

The management of reared tiger grouper (*Epinephelus fuscoguttatus*) infected with marine leech (*Zeylanicobdella arugamensis*)

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Tiger Grouper (*Epinephelus fuscoguttatus*) are the most cultured fish in Malaysia because of their taste and high prices. The culture of these fish provide a continuous supply domestically. Somehow, infestation of marine leech, *Zeylanicobdella arugamensis* as one of the crucial ectoparasitic diseases for marine fish especially grouper and could cause major problem in the culture industry. Health management of the fish included the monitoring of survival, feed intake, and growth rate (weight and length). SitroPro™, herbs developed by Fisheries Research Institute, Gelang Patah (FRI GP) located in the southern region of Peninsular Malaysia was used for the treatment of fish infected with parasites. The culture of Tiger Grouper (*Epinephelus fuscoguttatus*) grow steadily towards desired size resulting from good health management as well as good genetics which they inherit. Five families of tiger grouper raised in cages within the brackish water pond in FRI GP. The final survival rate for the families obtained were 87% (Johor♂ X Terengganu♀), 83.67% (Thailand♂ X Kedah♀), 72% (Indonesia♂ X Kedah♀) 72% (Kedah♂ X Johor♀) and 55.35% (Thailand♂ X Terengganu♀). A good health and water quality management with the support of appropriate treatment agent are very essentials for the health and survival of cultured tiger grouper (*Epinephelus fuscoguttatus*) in cages within the brackish water pond system.

Keywords: *Epinephelus fuscoguttatus*, SitroPro™, brackish water pond, water quality, *Zeylanicobdella arugamensis*

Short Biography

Mdm Fadzilah binti Yusof is currently working as Research Officer at the Fisheries Research Institute Gelang Patah, Johor, Malaysia. She completed her Bachelor of Science in Genetic from the University Kebangsaan Malaysia. She has 25 years experience in aquaculture since 1997 and have authored several publications in various journals and books. Her publications reflect her research interests in Aquaculture and Fish Health.

Preliminary study of water quality in relation to benthic organism under aquaculture cages situated in Rambungan River, Lundu

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Benthic organisms are commonly used as biology indicator for water quality as it differs in their tolerance towards pollution. For preliminary study, water quality data was collected at aquaculture cages situated at Rambungan River, Lundu for 6 months. Water quality parameter such as physical parameters (pH, temperature, dissolve oxygen dan salinity) are measured using multiparameter water quality probes (YSI Proplus) meanwhile biochemical parameters (ammonia, nitrate, nitrite dan phosphate) are analysed using spectrophotometer (HACH DR3900). Ekman Grab is used to collect benthic organism under the fish cages. The benthic organism was washed, separated and preserved in 7% buffered formalin and changed to 70% ethanol after 24 hour. From the study, it was found that the physical and biochemical parameters are relatively stable (within the water quality standard) for 6 months. The groups of benthic organism that are sampled are Crustacea, Annelids, Arthropod and Mollusca. As a conclusion, this study has to be continued and improved by gathering more data for a longer period of time and also relating it with the study of fish disease in cages.

Keywords: Benthic organism, water quality, aquaculture

Short Biography

Mdm Li Yung K is currently working as a research officer at the Fisheries Research Institute of Bintawa. She completed her degree on Science of Microbiology from Malaysia National University (UKM). She joined Fisheries Research Institute by 2012 in FRI Tanjung Demong and had experience working in shrimp processing factory before this. Currently she is working on the Project of Benthic Organism by relating it with water quality.

Dietary lacto-sacc improved growth performance and basic haematological parameters in empurau (*Tor tambroides*) fries reared in the aquaponics system

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Slow growth rate of empurau (*Tor tambroides*) fish is reported impeding its' aquaculture practice extensively in Malaysia. It mainly occurs at the fry to fingerling phase. Hence, the present research evaluated the dietary lacto-sacc feed supplement on the growth performance and health status of *T. tambroides* fries. The fish growth was outperformed significantly ($P<0.05$) after 12 weeks in fish fed with the lacto-sacc supplemented fish feed compared to the non-supplemented control diet. Alike the growth performance, the basic hematological parameters were significantly ($P<0.05$) improved. This study was the first attempt to indicate the suitability of dietary lacto-sacc for empurau fries nursery in the aquaponics system. Although this study was performed in four types of aquaponics systems, the factorial analysis confirmed that diet had a strong effect for this occurrence.

Keywords: Malaysian mahseer, *Tor tambroides*, growth, probiotics, lacto-sacc, hematology

Short Biography

Dr. Mohammad Bodrul Munir is currently working as senior lecturer at the Faculty of Resource Science & Technology, Universiti Malaysia Sarawak. He received his Doctoral degree or Ph.D on 2016 from the Universiti Sains Malaysia. Dr. Munir completed his Masters on 2003 from the Asian Institute of Technology, Thailand. He then worked at the international donor agency, served as Fisheries Management Officer at the DANIDA Technical Assistance Program. Dr. Munir has authored several publications in various journals and books. His publications reflect his research interests in fish nutrition particularly dietary prebiotics and probiotics and fish immunology.

Field evaluation of palm oil adjuvanted feed-based streptococcosis vaccine efficacy in red hybrid tilapia

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Vaccination plays an important role in the aquaculture industry to prevent disease outbreaks, thereby ensuring an economically viable industry. A field trial was conducted in Pedu Lake, Kedah, to determine immunoglobulin M (IgM) progression in red hybrid tilapia following oral vaccination with palm oil adjuvanted streptococcosis vaccine. A total of 6,000 tilapias of 60 ± 20 g were selected and divided into three groups of the single booster, double booster, and control group in duplicate. The single booster group was vaccinated orally at weeks 0 and 2, while fish of the double booster group were vaccinated at weeks 0, 2, and 6. Fishes from the control group were not given any vaccine throughout the experimental period. Samples of eye, brain, and kidney were collected at two weeks intervals for sixteen weeks cumulative to isolate the bacteria, while serum samples were collected to determine the antibody level. Throughout the study period, no outbreaks of streptococcosis were recorded, and none of the groups studied showed the presence of *Streptococcus agalactiae*. Very low mortality of single booster, double booster, and control group was noted at 2.1 ± 8.5 %, 1.3 ± 2.8 %, and 1.5 ± 7.1 %, respectively. Both vaccinated groups showed significant ($p < 0.05$) increases of IgM antibody levels following immunization of first and booster doses before went down below the cut-off value six weeks post-vaccination in the single booster group. However, IgM antibody levels in the fishes from the double booster group remained high until the end of the trial. In conclusion, oral vaccination with palm oil adjuvanted feed-based streptococcosis vaccine stimulates an adequate systemic immune response, which can confer possible protection against streptococcosis.

Keywords: Streptococcosis, *Streptococcus agalactiae*, Fish vaccine, Vaccination trial, Tilapia

Short Biography

Mr. Mohd Syafiq Mohammad Ridzuan is a research officer at National Fish Health Research Division (NaFish), Fisheries Research Institute (FRI) Batu Maung, Department of Fisheries Malaysia. His work focuses specifically on aquatic animal health research. He received a bachelor's degree in biomolecular science and is currently pursuing a master's in marine science at International Islamic University Malaysia.

Disinfection of rotifer, *Brachionus Plicatilis* leading to bacteria-free populations using SirehMAX™

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Rotifer is one of an essential food sources for marine fish larvae. Organic loads and microbial accumulation in rotifer cultures especially in batch culture are very high. Some of these bacteria can be pathogenic to rotifers. As rotifers are the first feed of fish larvae, their bacterial load can have a major impact on the survival. A reliable method was developed to disinfect rotifer, *Brachionus Plicatilis* by using SirehMAX™. SirehMAX™ is a herbal product containing betle leaves extract that has potential as an alternative to commercial antibiotic in aquaculture. The research was focused on determining the optimum exposure time and concentration dosage of SirehMAX™ for rotifer disinfection. The results indicate that the use of 10 ppm of SirehMAX™ on 400 ind/mL of rotifers for one hour is a very effective protocol for rotifer disinfection with negligible lethal effects on rotifers. The study has successfully tested disinfection protocols that were highly effective with regard to survival of rotifers.

Keywords: Rotifer, *Brachionus Plicatilis*, disinfection, SirehMAX™

Short Biography

Mrs. Fatin is currently working as Research Officer at the Fisheries Research Institute (FRI) Tanjung Demong, Besut Terengganu. She received her bachelor degree in Agriculture (Aquaculture) from the University of Putra, Serdang Malaysia.

Development of acquired immune system in striped catfish *Pangasianodon hypophthalmus*

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Striped catfish *Pangasianodon hypophthalmus* is one of the most important species in aquaculture in Vietnam. However, the mortality in the hatchery usually reaches around 70%-80% because of several infectious diseases. The understanding of the immune system, especially acquired immune system, is important to improve the survival rate. In this study, tissue distribution of genes related to acquired immunity such as CD8, TCR and rag1 were investigated in this fish species. Further, the gene expressions of these genes in each developmental stage of the larvae were also studied to reveal appearance of the acquired immune system during the development process. The brain, fin, gills, head kidney, intestine, liver, muscle, skin, spleen, thymus and trunk kidney from four fingerlings ($22.8 \pm 1.8\text{g}$) were collected in nucleic acid preservation buffer. Further, the larvae at 2 days post-hatchery (dph), 3 dph, 5 dph and 10 dph were also collected as above. Quantitative RT-PCR for CD8, TCR and rag1 were performed using cDNA samples from each tissue or larvae sample. Transcripts of CD8, TCR and rag1 gene were detected in all tissues examined, while the highest levels for all genes were observed in the thymus. CD8, TCR and rag1 transcripts were detected from the larvae sampled at 2 dph, 3 dph and 5 dph. These data provide basic knowledge of T-cell related genes in *P. hypophthalmus* and suggest that T cell development have been already started at 2 days post-hatchery in this fish species.

Keywords: striped catfish, *Pangasianodon hypophthalmus*, larva, acquired immune genes, gene expression

Short Biography

Nguyen Ngoc Dung is currently working as a lecturer at Can Tho University, Viet Nam. She is a third year doctoral student at Marine Biosciences from Tokyo University of Marine Science and Technology, Japan. She completed her master of Fisheries Science from Nagasaki University, Japan. Her research focuses on fish immunology.

DAY 4: 26 August 2022 (Friday)
Session 6: Trends in Shrimp Health Management



Keynote 6

The quorum sensing (QS) system modulates virulence of *Vibrio parahaemolyticus*^{AHPND} by regulating the expression of the *pirA*^{vp}/*pirB*^{vp} genes through AphB^{vp}

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The AHPND-causing *Vibrio parahaemolyticus* strain, the *pirA*^{vp} and *pirB*^{vp} genes are highly expressed in the early log phase of the growth curve, and that expression of the PirA^{vp} and PirB^{vp} proteins continues throughout the log phase. In this study, when we compared mutant strains with a deletion or substitution in two of the quorum sensing (QS) master regulators, *luxO* and/or *opaR* (*luxO*^{D47E}, Δ *opaR*, Δ *luxO* and Δ *opaR* Δ *luxO*), our results suggested that expression of the *pirA*^{vp} and *pirB*^{vp} genes was related to the QS system, with *luxO* acting as a negative regulator of *pirA*^{vp} and *pirB*^{vp} without any mediation by *opaR*. We also identified a putative consensus AphB binding site in the promoter region of *pirAB*, and used an electrophoretic mobility shift assay (EMSA) to show that AphB^{vp}, but not AphA^{vp}, could bind to this predicted region. Real-time PCR further showed that *aphB*^{vp} was negatively controlled by LuxO^{vp}, and that its expression was parallel to the expression patterns of *pirA*^{vp} and *pirB*^{vp}. Taken together, these findings suggest that the QS system may regulate *pirA*^{vp}/*pirB*^{vp} expression through AphB^{vp}. This new insight into the pathogenic mechanisms of AHPND points toward the QS system as a possible target for therapeutics that might one day be able to control the virulence of AHPND-causing bacteria and prevent AHPND.

Keywords: shrimp, AHPND-causing *Vibrio parahaemolyticus* strain, quorum sensing (QS) system, AHPND pathogenicity

Short Biography

Dr. Lo is the Emeritus Chair Professor of National Cheng Kung University, Taiwan. Dr. Lo's research work not only focuses on shrimp white spot disease (WSD) but also a shrimp bacterial disease acute hepatopancreatic necrosis disease (AHPND), caused by a pathogenic strain of the opportunistic marine bacterium *Vibrio parahaemolyticus*. Dr. Lo's team has constructed the mutants of *V. parahaemolyticus*^{AHPND}, and successfully demonstrated the pathogenic mechanisms of AHPND from many aspects. These new insights into the pathogenic mechanisms of AHPND provide important information for developing effective AHPND control measures.

Oral presentation

***Piper betle* L. leaf extracts inhibit quorum sensing of shrimp pathogen *Vibrio harveyi* and protect *Penaeus vannamei* postlarvae against bacterial infection**

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Vibriosis, with *Vibrio harveyi* as one of its causative agents, is a major disease in shrimp causing huge economic losses in the Philippines, Thailand, Vietnam and other Southeast Asian countries. It is induced through the formation of biofilm as a result of bacterial cell-to-cell communication or quorum sensing (QS). Hence, this mechanism may be targeted in controlling *V. harveyi* infections in shrimp. In this study, crude extract (CE) and crude alkaloids (CA) from Ikmo (*Piper betle* L.), a plant native to Southeast Asia, were observed to significantly ($p < 0.05$) inhibit biofilm formation in chitosan-coated microtiter plates of wild-type strains *V. harveyi* VH0, VH1 and BAA-1116 without inhibiting their growth. Confocal laser scanning microscopy showed thinner biofilms formed upon treatment with the extracts. Furthermore, both CE and CA significantly ($p < 0.05$) inhibited bioluminescence in *V. harveyi* BAA-1116 and was shown to interfere with QS by modulating autoinducer (AI) activities as observed in both phenotypic and gene expression analyses. Both extracts also did not negatively affect shrimp growth and pre-infection mortality rate. Despite the *in vitro* results however, *in vivo* analysis showed that only *P. betle* CE protected *Penaeus vannamei* postlarvae against *V. harveyi* infection after seven days. These show the potential supplementation of shrimp feed with *P. betle* crude extract as protection against AHPND.

Keywords: Biofilm inhibition, Feed supplementation, *Piper betle*, Quorum sensing inhibition, *Vibrio harveyi*

Short Biography

Mr. John Paul Matthew D. Guzman is currently working as a Science Research Analyst at the Industrial Technology Development Institute, Department of Science and Technology, Philippines, and a lecturer at the *Pamantasan ng Lungsod ng Maynila* (University of the City of Manila), Philippines. He received his Master of Science degree in Microbiology from the University of Santo Tomas. Mr. Guzman completed his Bachelor of Science in Biology from the *Pamantasan ng Lungsod ng Maynila*. Mr. Guzman has authored a number of publications in various journals. His publications reflect his research interest in microbial control, particularly via quorum sensing inhibition.

Involvement of White Spot Syndrome Virus viral proteins in modulating shrimp glycolysis

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White spot syndrome virus (WSSV) has been a critical issue in the shrimp-farming industry, causing enormous economic losses. To control this viral disease, the molecular mechanism underlying WSSV pathogenesis must be well understood. WSSV triggers the glycolytic pathway to benefit viral replication, but this regulation is not well elucidated. We determined that the activity of glycolytic enzymes, namely Phosphofructokinase (PFK), Lactate dehydrogenase (LDH), and Pyruvate kinase (PK), were increased at the viral genome replication stage (12 hpi). Silencing of glycolytic enzymes and glycolysis disruption by 2-DG indicated WSSV has a heavy reliance on glycolysis to achieve successful replication. We also demonstrated that the gene expression of LDH was not increased at 12 hpi, even though its activity was increased. This prompted us to investigate whether regulation of LDH involved an interaction between viral protein and LDH. With Yeast 2-hybrid studies, we determined that LDH interacted with two WSSV proteins, WSSV ORF A and WSSV ORF B. The interaction between viral protein and LDH was further confirmed in Sf9 cells transfection. Interestingly, the interaction with WSSV ORF A did not increase LDH activity in either Sf9 or shrimp hemocytes. Furthermore, WSSV ORF A silencing increased LDH activity and impaired virus replication. Taken together, this study demonstrated viral protein from WSSV can interact with LDH. WSSV ORF A may have an inhibitory role in regulating LDH activity, with potential as a strategy to impede virus spread.

Keywords: White Spot Syndrome Virus, White Shrimp, Glycolysis, Lactate dehydrogenase, Viral protein

Short Biography

Mr. Yen Siong Ng is a PhD student at the National Cheng Kung University. He received his MS in 2020 from the same university. His research is focused on how white spot syndrome virus (WSSV) regulates glycolysis in shrimp to support viral replication. He has attended various conferences and was awarded first prize at The Metabolomic Analysis and Application Conference and 1st runner up for a poster presentation at the Fisheries Society of Taiwan conference, both in 2022.

Shrimp GDH may be regulated by SIRT4 during WSSV replication

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White spot disease (WSD), caused by the white spot syndrome virus (WSSV), inflicts economic losses in shrimp aquaculture. To support viral replication, WSSV induces metabolic rerouting in shrimp, including reductive glutamine metabolism. WSSV induces glutamate dehydrogenase (GDH) expression at the replication stage; it is an enzyme involved in reductive glutamine metabolism, but regulation of host factors and viral proteins are not well understood. In mammalian research, GDH is inhibited by SIRT4, a NAD-dependent ADP-ribosyltransferase, member of sirtuins(SIRT) family. It may be important in metabolic rerouting in cancers or in virus-infected vertebrate cells. Although SIRT4 has been widely studied regarding regulation of glutamine, SIRT4 has not been characterized in *L. vannamei*. Our objective was to determine involvement of LvSIRT4 in WSSV-induced glutamine metabolism. First, LvSIRT4 was identified and characterized and recombinant LvSIRT4 and LvGDH were localized by immunofluorescence in insect Sf9 cells. Then we clarified the relation between GDH and SIRT4 by using Co-IP and GDH enzyme activity detection. Surprisingly, GDH enzyme activity was increased in LvSIRT4 silencing or overexpress shrimp during WSSV infection, but the increase of LvSIRT4 silencing shrimp was slower than LvSIRT4 overexpression in shrimp. When potential involvement of WSSV viral proteins with LvSIRT4 as well as these enzymes was assessed using Co-IP, there were indications that shrimp SIRT4 regulated GDH activity independent of their interaction. Furthermore, we confirmed that mRNA expression of LvSIRT4 was increased during WSSV infection and silencing of SIRT4 with dsRNAs decreased WSSV gene expression and genome copy numbers. Overexpression of LvSIRT4 increased WSSV gene expression and genome copy numbers. Based on our current data, we inferred that LvSIRT4 promoted GDH enzymes activity and WSSV replication in shrimp. In the near future, we plan to elucidate the regulatory mechanism of LvSIRT4 increased GDH enzyme activity and further characterize the SIRT4-mediated mechanism in WSSV-induced glutamine metabolism.

Keywords: SIRT4, white spot syndrome virus (WSSV), glutamine metabolism

Short Biography

Tan Yu Kent is currently a MSc student in the Department of Biotechnology and Bioindustry Sciences, National Cheng Kung University. In the past two years, his research focus has been the pathogenesis of shrimp white spot syndrome virus.

WSSV-induced metabolic reprogramming in shrimp hemocytes to benefit its replication

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Shrimp farming is a fast-growing sector of aquaculture that aims to meet the growing global food demand. Outbreak of white spot disease (WSD) caused by white spot syndrome virus (WSSV) and mass mortality remains a threat to the industry. The molecular pathogenesis of WSD is not well understood. In the last few years, advances in omics technology have led to the understanding of the WSSV-induced metabolic reprogramming in shrimps, including aerobic glycolysis and glutamine metabolism. Using multi-omics approach, several shrimp and WSSV databases were established which serves as a fundamental base for our ongoing research. Combining omics data with stable isotope tracking resulted in identifying key shrimp metabolic and signaling pathways hijacked by WSSV. Given the daunting complexity of the host-pathogen interactions, we anticipate that our current findings will be a framework to understand the complex WSSV-WSSV and shrimp-WSSV protein-protein interactions. New insights gained from the comprehensive interactome databases and collaborative research efforts would increase our understanding of the dynamic host-pathogen interactions and facilitate the rational development of effective strategies to mitigate the viral replication.

Keywords: shrimp, White spot syndrome virus, aerobic glycolysis, glutamine metabolism

Short Biography

Dr. Wang is currently a Distinguished Professor and also holds the University Library Curator position at National Cheng Kung University, Taiwan. Dr. Wang uses an integrated systems-biology and omics approach, including metabolomics and lipodomics, to study the complex biological phenomena of WSD and AHPND pathogenesis and to develop disease control strategies that are effective and eco-friendly. She always looks for novel ways to apply scientific findings to address the problems of shrimp diseases; consequently, the shrimp aquaculture industry has greatly benefitted from using the new knowledge that she has generated to develop evidence-based biosecurity management plans.

Quorum quenching activity in microalgae and its associated bacteria: an alternative antimicrobial therapeutics

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Disease outbreaks are one of the major drawback that hinder the sustainable development of aquaculture production. The application of synthetic chemicals and antibiotics to overcome diseases in aquaculture has achieved partial success. However, the misuse of antibiotics has led to the development of antimicrobial resistance (AMR) microorganisms that impose a serious threat to the public health. Hence, alternative approach such as application of the potential indigenous microalgae and its associated bacteria could act as a consortium to control diseases caused by bacteria in aquaculture. One of the control mechanism identified in the potential microalgae and its associated bacteria is quorum quenching (QQ) –the disruption of the cell-to-cell communication system of the bacteria. The cell-to-cell communication of the bacteria is known as quorum sensing (QS). In response to population density, QS regulates virulence factor of bacteria through the production of small signal molecules. In this study, the QQ activities of different marine microalgae and their associated bacteria were evaluated *in vitro* using QS biosensors and *in vivo* using *Artemia franciscana* as a model organism. Among the 17 microalgae species tested, a local strain of green alga, identified as *Chlorella* sp. inhibited purple production of *Chromobacterium violaceum* CV026 (without inhibiting the biosensor's growth), reduced bioluminescence of *Vibrio campbellii* BB120 and its mutant strain of *V. campbellii* JMH597 as well as improved *Artemia* survival when challenged with *V. campbellii* BB120.

Keywords: Quorum quenching (QQ); Quorum sensing (QS); Antimicrobial resistance (AMR); *in vitro* and *in vivo*

Short Biography

Dr. Sarmila is a postdoctoral researcher at the Universiti Putra Malaysia (UPM). She received her PhD in Aquaculture from the Universiti Putra Malaysia (UPM). Dr Sarmila completed her Masters in Microbial Biotechnology from the University of Malaya (UM). She then worked at Biovalence Sdn.Bhd for the past 3 years as a Head of Shrimp Disease Centre (SDC). Dr. Sarmila has authored several publications in various journals and books. Her publications reflect her research interests in shrimp diseases, genomic, quorum sensing and probiotics.

Microbiome signatures from stocking to emergency harvest in Thailand *Penaeus vannamei* pond Experienced Yellow Head Virus outbreak

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Altered microbiome structures in animals have been evidently observed in diseased or syndromic conditions, so called pathobiome. Shrimp intestinal and rearing water microbiomes have been reportedly perturbed in various viral and bacterial outbreaks, but transition from healthy microbiomes to pathobiomes in a yellow head virus (YHV) outbreak aquacultural pond has not been investigated. Here, we examined transitional microbiomes of shrimp intestine and rearing water samples from healthy to emergency harvest for a complete eight-week culture period of a Thailand *Penaeus vannamei* grow-out commercial pond experienced shrimp mortality and YHV infection. Each week from postlarvae stocking to the emergency harvest, 10 shrimp intestine and four pond-corner-water samples were collected. Microbiomes in the collected samples were assessed by amplicon sequence variants (ASVs) produced with QIIME2 on Illumina MiSeq high-throughput V3-V4 16S rRNA amplicon sequencing reads of extracted total DNA. ASVs were assessed for taxonomic classification, alpha and beta diversity indices and differentially abundant ASVs with DADA2, CoDA and phyloseq packages in R, in conjunction with rearing water quality parameters and pathogen detection PCR results. Water and shrimp intestinal bacterial profiles shifted from the stocking to the emergency harvest in principle component analyses. Increasing heterogeneous bacterial profiles in the intestines were observed from weeks 6 to 8, and intestinal and water bacterial profiles moved closer to one another in week 8. The increased heterogeneity and altered alpha diversities of the intestinal and water bacterial profiles may be associated with increased YHV loads detected in week-7 water and later in week-8 water and shrimp. We also examined characteristics and profiles of ASVs as potential early-warning taxa candidates of YHV outbreaks. Our analyses revealed dynamic and transitional bacterial structures in the *P. vannamei* pond experienced YHV emergency harvest, warranting further characterization and validation of YHV pathobiomes and potential early-warning signals.

Keywords: microbiome, pathobiome, *Penaeus vannamei*, yellow head virus (YHV), early-warning signals

Short Biography

Dr Prachumwat received his doctorate in Genetics from University of Chicago, USA. He is interested in utilizing genomic sequences to better understand interactions between shrimp, their pathogens and microbiomes. He analyzes genomes and transcriptomes of several shrimp pathogens for developing detection methods and strategies to control infections and reduce virulence. He and his colleagues investigate microorganisms and pathobiomes of shrimp aquaculture systems for shrimp health and pathogen discovery of current and potential future emerging shrimp diseases

Dietary montmorillonite clay protected *Penaeus vannamei* challenged with *Vibrio parahaemolyticus* (AHPND) and modulated stomach microbiota of shrimp survivors

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Acute Hepatopancreatic Necrosis Disease (AHPND) is currently causing a bacterial pandemic in the shrimp farming world. AHPND is now known to be caused by pathogenic strains of *Vibrio* bacteria that produces PirAB-like toxins. Shrimp exposure to AHPND-causing bacteria is through the oral route, ingested into the digestive tract where the bacteria initially colonized the stomach and then releases the binary toxins to damage the hepatopancreas. It has been reported that *Vibrio parahaemolyticus* strains causing AHPND (VP_{AHPND}) has now shown antibiotic resistance. There is currently much interest in the development of functional aquafeeds containing additives such as prebiotics, probiotics, organic acids and phytogenics. Another alternative to mitigate AHPND is to bind the bacterial toxins released by adding adsorbent clays into shrimp feeds. A feeding trial using triplicate groups of 20 SPF shrimp per replicate was conducted to evaluate the efficacy of a montmorillonite clay, CL (Calibrin[®]-Z, Amlan International, USA) on *Penaeus vannamei* postlarvae growth, gut health and disease resistance to AHPND. The addition of 0.25% or 0.50% CL did not significantly ($P>0.05$) impact growth and feed utilization efficiency. When challenged with a dose of 10^8 CFU/mL VP_{AHPND}, survival of shrimp fed 0.25%CL or 0.50%CL were 83.3% and 93.8%, respectively, and were significantly higher ($P<0.05$) compared to the positive control group which registered a survival of only 39.6%. *Vibrio* and total cultivable bacteria counts in the hepatopancreas of shrimp fed CL-added diets were significantly lower compared to positive control. Histopathology of the hepato-pancreas of infected shrimp fed CL-added diets showed less damage compared to the positive control group. Dietary CL enhanced stomach microbiota diversity and seemed to have modulated the bacterial community possibly with beneficial impact to shrimp survival. Specialized clays can be used as functional feed additives in shrimp feeds for AHPND mitigation.

Keywords: Montmorillonite clay, *Penaeus vannamei*, AHPND, microbiota, functional feeds

Short Biography

Dr. Wing-Keong Ng obtained his Master's degree in Aquaculture from the Asian Institute of Technology (Thailand), and doctorate in Nutrition at the University of California (Davis, USA), followed by a post-doctoral fellowship at Mississippi State University. He was a senior professor at Universiti Sains Malaysia and is currently an independent aquafeed consultant. A recent study led by researchers from Stanford University ranked Dr. Ng among the world's top 2% scientists based on career-wide scientific impact across disciplines. He is on the editorial board of several international journals and a consultant to various international aquaculture organizations and aquafeed-related companies.

3 Minutes Presentation

The effectiveness of probiotic used in super intensive culture of white shrimp (*Penaeus vannamei*) for its health and water quality management

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The study aimed to evaluate the effect of a commercial probiotic applied in super intensive shrimp culture system without stopping the good water quality as well as the shrimp good health. Probiotics is globally known as “bio-friendly agents”, can be introduced into culture environment to control the optimum water quality ultimately to stimulate a better growth of the cultured organism besides its believed to compete with pathogenic bacteria. The super intensive culture system manoeuvres to strike a balance between high shrimp productivity, maintained good water quality although in a very minimal water exchange condition. The glitches that are frequently stumble upon super intensive shrimp culture system (*Penaeus vannamei*) are diseases occurrence and degradation of the water quality parameters. Probiotic supplement is one of the most important management strategies to increase shrimp growth in a super intensive system with minimal water exchange. Results showed that with the addition of the probiotic chosen, concentrations of ammonia, nitrate, nitrite and phosphorus were reduced, and increased the yields of shrimp. It was also showed a significantly increased dissolved oxygen ($P < 0.05$) besides reduced dissolved reactive-phosphorus, total inorganic nitrogen and biological oxygen demand ($P < 0.05$). Three cycles of pacific white shrimp culture were done with stocking density (SD) of 100PI/m² (control), 250PI/m² and 300PI/m² for 120 days per cycle. Total shrimp yield for both SD (250PI/m² and 300PI/m²) were significantly higher ($P < 0.05$) than SD 100PI/m² which was 41.0mt/ha, 46.0mt/ha and 11mt/ha of marketable sized shrimp respectively. Those indicated that the addition of the commercial probiotics had a noticeable influence on water quality of shrimp ponds and shrimp health which derived higher shrimp production.

Keywords: *Penaeus vannamei*; Super-intensive culture system; probiotic; water quality; minimal water exchange

Short Biography

Azmi bin Rani is currently working as Research Officer at the Fisheries Research Institute Gelang Patah, Johor, Malaysia. He completed his Bachelor of Science in Biotechnology from the Universiti Putra Malaysia (UPM) . Aquaculture experience since 1997 especially in Marine Fish and Shimp culture and have authored several publications in various journals and books. His publications reflect his research interests in Aquaculture and Fisheries Resources.

Preliminary study on the effect of moist formulated feed on health status of disease infected *Penaeus monodon* Broodstock

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Currently, most all of shrimp (*Penaeid* sp.) hatcheries still rely on fresh feed such as squid, fish, polychaete and bivalves as broodstock feed. However, they are directly exposed to the risk of disease infection such as parasites, bacteria and viruses from fresh feed even after being frozen. Therefore, a disease-free moist formulated feed for shrimp broodstock has been developed and preliminarily tested to evaluate the effect of the use of moist formulated feed (MFF) compared to fresh feed on the health status of tiger shrimp, *Penaeus monodon* broodstock. Under this study, a total of 30 pairs of wild tiger shrimps (*P. monodon*) with averaged weight of $64.3 \pm 15.2\text{g}$ (♂) and $103.9 \pm 13.4\text{g}$ (♀) were stocked in two blocks of three 5MT tanks each at a stocking density of 5 pairs/tank with 1:1 sex ratio. Preliminary disease screening with RT-PCR found that 50% of the samples were infected with IHHNV and 80% of the samples were infected with WSSV. The first block of shrimp broodstock were fed with fresh squid and commercial pellets (D1) and the second block were fed with MFF at a rate of 7.5%/day of biomass weight (0930 and 1630 hours). Wilcoxon analysis comparing the IHHNV and WSSV virus presence scores for 24 weeks showed that all tiger shrimp broodstock were found to be free of IHHNV and WSSV infection ($Z = 2.558$, $p = 0.011$) compared to week -1. While the analysis of Independent Samples T-Test on survival rate (average \pm SD%) showed no significant difference of tiger shrimp broodstock fed D1 ($60.0 \pm 15.5\%$) and MFF ($50.0 \pm 17.9\%$) in condition; $t(10) = 1.04$, $p = 0.325$. The results suggest that MFF could be used as an alternative feed for tiger shrimp broodstock.

Keywords: shrimp broodstock, *Penaeus monodon*, disease infection, moist formulated feed

Short Biography

Mohammed Suhaimee Abd Manaf is currently working as Director at Fisheries Research Institute Pulau Sayak, Kedah, Department of Fisheries Malaysia. He received his Master degree in Chemistry from the University of Technology, Malaysia. He completed his BSc on Biochemistry and Microbiology from the University of Sheffield, England. He then worked at the Department of Fisheries Malaysia, served as a Fish Nutritionist at the Brackishwater Aquaculture Research Centre, Gelang Patah, Johor, Malaysia. He has authored several publications in various journals and books. His publications reflect his research interests in fish and nutrition.

Blue-green algae and assessment of microcystin in shrimp aquaculture farms of Sarawak

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Blue-green algae blooms can cause severe water quality deterioration including scum formation and toxin production. A total of 20 small scale shrimp farms were assessed from February 2021 to November 2021 for the abundance of blue-green algae (cyanobacteria) and the levels of microcystin in the tissue of shrimps using enzyme-linked immunosorbent assay (ELISA). There was a high cell count of *Microcystis* sp at 6.77×10^8 cells/ L in Muara Tebas, *Anabaena* sp at 4.99×10^7 cells / L in Telaga Air and *Pseudanabaena* sp at 1.69×10^8 cells/ L in Kuala Baram. Microcystin was detected but at a low level in the shrimp samples collected from the shrimp farms in Sarawak throughout the study. This study demonstrated that blue-green algae monitoring in shrimps aquaculture farms is necessary.

Keywords: Blue green algae, microcystin, ELISA, shrimps, aquaculture

Short Biography

Ms Lim Mui Hua is currently working as a Research Officer at Fisheries Research Institute Bintawa, Sarawak. She received her Bachelor's and Master's degree from UNIMAS. She has over 19 years of experience in the field of research in Fisheries Biotechnology especially on the detection of biotoxins and study of microalgae. She has published many papers in various journals

Scientometrics of climate change and shrimp disease: An overview

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As the FAO fishery statistic, the total global production of shrimp is in increasing trends, and shrimp has been considered as one of the crucial components of the coastal fisheries resources in many countries, especially in Asia. Sea surface temperature, sea level rise, coastal flood, coastal erosion, ocean heatwave and ocean acidity are among multiple climatic impact-drivers that are projected to change in coastal and open oceans regions of the world, especially when most of the aquaculture activities are actively being carried out in the areas. Thus, the aim of this study is to synthesize the available literature on the impacts of the climate change on shrimp disease in Asia using the Scientometric method. We first identified the top disease in shrimp aquaculture along with climatic impact-drivers of climate change, then extracted bibliometric from Web of Science. Then we used CiteSpace to assess trends and status of research focusing on the impacts of climate change on worldwide shrimp disease. To address gaps in this emerging field, we make two main recommendations: (i) Increased collaboration between countries to develop global solutions towards shrimp disease in the face of climate change and (ii) Drawing from other disciplines outside of “Ecology, Earth, Marine”, such as molecular biology, economics, and sustainability, would strengthen and provide additional insights on sustainable shrimp production in the world. Further qualitative discussion was provided along with the future research topic’s direction of Sustainable Development Goals of Food Security and Climate Change.

Keywords: Aquaculture, Environmental Science, Global Warming, White spot syndrome virus, whiteleg shrimp

Short Biography

Dr. Mohamad Nor Azra is a Senior Researcher and Head of Climate Change Adaptation Laboratory, Institute of Marine Biotechnology, Universiti Malaysia Terengganu and among one of the Inaugural Fellow for the Leadership for Climate Resilient Fisheries, U.S.A. He was the project coordinator for invasive species relates to the environmental sustainability and societal wellbeing, as well as the Malaysian Long-Term Research Grant Scheme for climate change impacts towards the aquatic ecosystem and member in other research international projects or grants. He is author and co-author of 60 scientific papers, two book, one edited book, and three book chapter. His research areas include: 1) Effects of climate change on marine aquatic organism 2) invasive species towards sustainable environment and societal wellbeing 3) Physiological and behavioral changes of the crustacean

Microsporidian parasite, *Enterocytozoon hepatopenaei* at early stage of culture of whiteleg shrimps (*Peneaus vannamei*) in West Malaysia

Rohaiza Asmini Yahya*, Padilah Bakar, Wan Muhammad Hazim Wan Sajiri, Nur Samihah Mazlan, Nur Ashikin Arbi and Kua Beng Chu

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The hepatopancreatic microsporidian *Enterocytozoon hepatopenaei* (EHP) is an emerging pathogen that affects cultured shrimp. Shrimp infected with EHP showed a clinical sign of stunted growth as early as 14 days in the culture system. The objective of this study is to determine the current status of EHP infestation and its prevalence in the early-stage culture of whiteleg shrimp (*Penaeus vannamei*). A cross-sectional study was conducted at selected farms in Terengganu, Pahang, Johor and Selangor as early as 14 days in culture system and less than 30 days of culture in the pond. The detection of EHP gene sequence from hepatopancreas tissues of shrimps was determined using Polymerase Chain Reaction (PCR). Unique spore wall protein gene-specific (SWP) for EHP was used in this diagnosis, which was infected hepatopancreatic tissue. Our study showed high prevalence of EHP in Pahang (100%), Terengganu (96.7%), Selangor (73.3%) and Johor (26.7%). EHP infestation were contributed by many factors, including the source of larvae, control of vectors as potential carriers and disease transmissions, biosecurity and hygienic practices in daily routine farm management, water exchange and water quality as well as pond management.

Keywords: *Enterocytozoon hepatopenaei*, parasite, prevalence, spore wall protein, whiteleg shrimps

Short Biography

Rohaiza Asmini is currently working as researcher at the National Fish Health Research Division (NaFisH), Fisheries Research Institute (FRI) Batu Maung, Pulau Pinang, Malaysia. She received her Bachelor's degree in Applied Science (Fisheries) from the Universiti Malaysia Terengganu. She has authored several publications in various journals and newsletters. Her publications reflect his research interests in parasitology on aquatic animals.

Unveiling the potential of medicinal plants in combating shrimp acute hepatopancreatic necrosis disease (AHPND)

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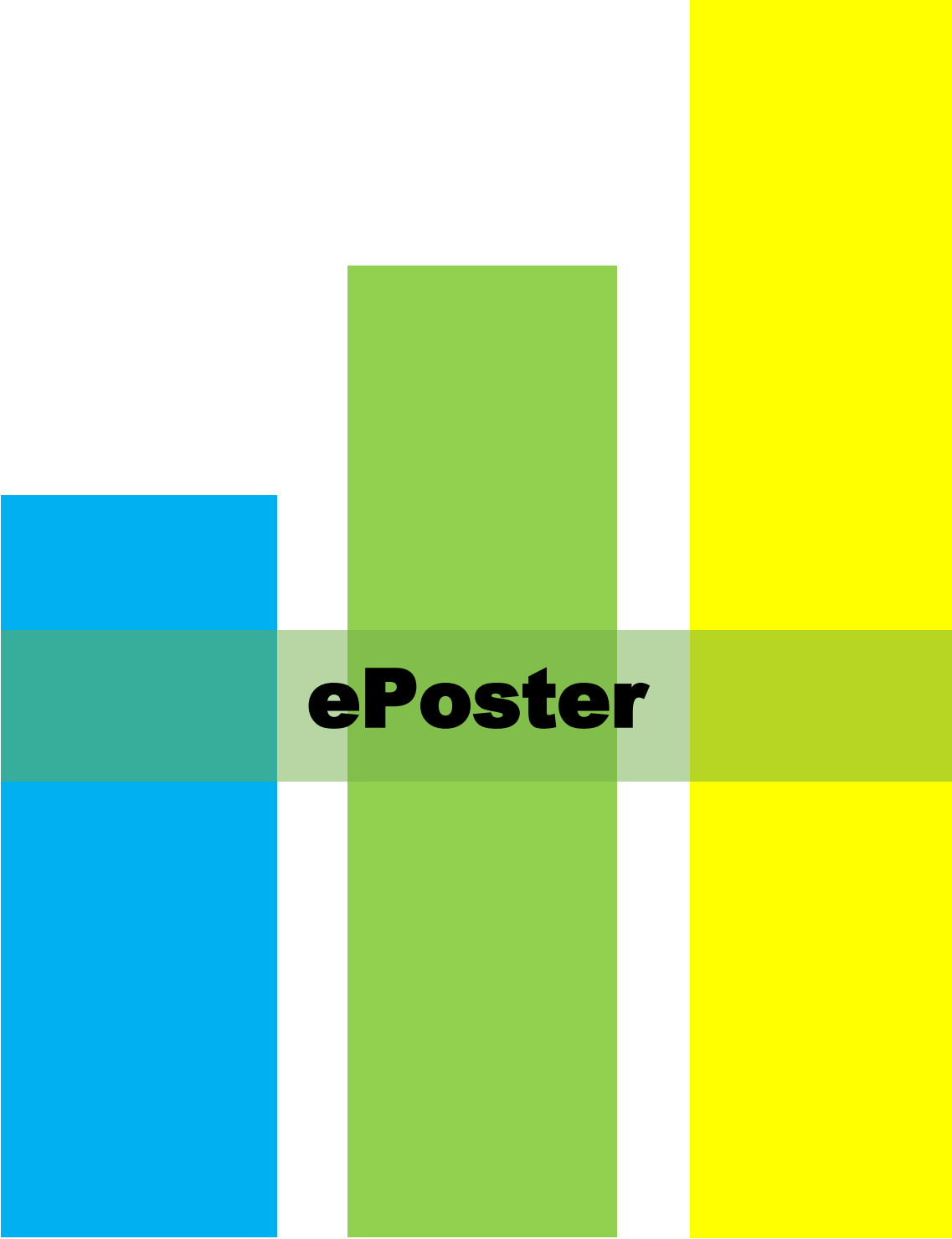
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Acute hepatopancreas necrosis (AHPND) is an emerging disease that causes tremendous economic losses to the shrimp-farming industry worldwide. The causative agent of AHPND is an opportunistic pathogen called *Vibrio parahaemolyticus*. The virulence repertoire is granted by the possession of a lethal binary toxins PirA/PirB, which attacks the hepatopancreatic cells of shrimps. The hitherto lack of a specific antibacterial strategy or control measurement have resulted in spontaneous infections that significantly reduce the shrimp production. In an attempt of screening on 85 common medicinal plants that would mitigate the pathogenicity of *V. parahaemolyticus* the aqueous extracts of two plants (CTM27 & CTM50) were found to specifically inhibit the growth of the pathogen as shown by zone inhibition studies. The two extracts inhibited the growth of *V. parahaemolyticus* at a relatively minimal concentration of 100 mg/ml (dried plant sample/volume). As the biggest surprise, another two plants' extracts (CTM15 & CTM44) which at same concentrations were able to neutralize the antagonism activity of *V. parahaemolyticus*. Similar with other closely related pathogens, *V. parahaemolyticus* confers antagonism activity that gains enhanced fitness by secreting effector proteins to prevent the growth of other competitive bacteria living within the same habitat. The two extracts reversed the antagonism activity of *V. parahaemolyticus* against *Escherichia coli* as deduced from the preliminary laboratory results. Collectively, this study discovers that two plants' extracts inhibit the growth of *V. parahaemolyticus*; and two plants reverse the antagonism activity of *V. parahaemolyticus*. They are ideal candidates to be developed into a disease control diet owing their heat stability and relatively lower cost.

Keywords: AHPND, shrimp, medicinal plant, antagonism.

Short Biography

Ms. Tan Wen Chian is currently working as a Ph.D. student at UCSI University. She received her MSc Applied Sciences by Research from the same university."



SESSION 1: BIOSECURITY IN AQUACULTURE

Economic Impact of Vibrio Vaccination on Cage-Cultured of Asian Seabass (*Lates calcarifer*)

Arbania binti Ali, Jumria binti Sutra, Ina Salwany Md. Yassin, Mohammad Noor Amal Azmaid, Siti Hajar Mohd Yazid and Norhariyani Mohd Nor

Biofloc composition in Whiteleg shrimp, *Penaeus vannamei* culture system as biosecurity approach towards sustainable shrimp aquaculture production

Hidayah Manan, Mhd Ikhwanuddin, Fazlan Fauzan, Ashraf Suloma and Nor Azman Kasan

Fatty acid compositions and oil quality assessment of liver of Yellow puffer, *Xenopterus naritus*

Samsur Mohamad, Nur Eqmal Dinie Nor Azmi and Zaini Assim

Advances in technologies towards enhancing shellfish wellbeing for optimum aquaculture production

Benedict Terkula Iber, Hidayah Manan, Muhammad Syafiq Abd Razak, Hassimi Abu Hassan and Nor Azman Kasan

Sustainable Fishery and Aquaculture Development of Yellow Pufferfish from Sarawak, Malaysian Borneo

Ahmad Syafiq Ahmad Nasir, Samsur Mohamad and Mohammed Mohidin

Gamma ray irradiation: A Valuable tool for fresh feed disinfection

Pik Neng Teoh, Che Zulkifli Che Ismail, Nor Aida Suzana Abdul Rahman and Nazaria Mohd Nazri

Evaluating antibiotic susceptibility profile of Vibrio species isolated from cultured Epinephelus species in Peninsular Malaysia

Ina-Salwany Md Yasin

SESSION 2: EPIDEMIOLOGY

Parasite Infestation in *Macrobrachium rosenbergii* from Sarawak (Kuching-Samarahan Region)

Rizmanisha-Jasmin Ruzaiuddin and Fatimah-A'tirah Mohamad

Shrimp with positive PCR for DIV1-ATPase gene revealed no pathological lesions of DIV1

Rungkarn Suebsing, Piyachat Sanguanrut, Sukanya Jitchana, Somjai Wongtripop, Timothy W. Flegel and Kallaya Sritunyalucksana

First report of Sacculinidae parasite infestation on crab population (Subfamily Thalamitinae) in Peninsular Malaysia

Hanafiah Fazhan, Mhd Ikhwanuddin and Khor Waiho

SESSION 3: DETECTION METHOD/DIAGNOSTIC (PARASITIC, BACTERIAL & VIRAL DISEASES)

The externa histological description of a sacculinid (Crustacea; Rhizocephala) and its impact on the gonadal development of mud crab

Khor Waiho, Hanafiah Fazhan, Henrik Glenner, Julia Hwei Zhong Moh and Mhd Ikhwanuddin

Microbiome analysis of gut bacterial communities of healthy and diseased Malaysian mahseer (*Tor tambroides*) using 16S rRNA metagenomics approach

Melinda Mei Lin Lau, Cindy Jia Yung Kho, Leonard Whye Kit Lim, Siao Chuang Sia, Hung Hui Chung, Samuel Lihan and Kasing Apun

Pathogenicity of Different Betanodavirus Strains in Asian Seabass (*Lates Calcarifer*) Under Temperature Fluctuation Stress

Azila, A., Safwan, M. K. A., Firdaus-Nawi, M., Shaharah, M. I., Rimatulhana, R. and M Nur-Nazifah

Improvement of a method for detecting *Perkinsus olseni* in low infection intensity samples

Vui Kien Liew and Tomoyoshi Yoshinaga

SESSION 4: PREVENTION & CONTROL MEASURES (PARASITIC, BACTERIAL & VIRAL DISEASES)

Production of biochar via microwave pyrolysis of palm kernel shell for simultaneous removal of ammonia and cultivation of lettuce in aquaponic system

Su Shiung Lam, Wan Adibah Wan Maharia, Man Huan Sua and Elfina Azwara

Immune response and tolerance of White-leg shrimp *Penaeus vannamei* to *Vibrio parahaemolyticus* following exposure to *Pandanus tectorius* leaf extract

Yeong Yik Sung, Anupa Anirudhana and Yosie Andriana

Effect of dietary prebiotics on snakehead (*Channa striata*) health: haematology and disease resistance parameters against *Aeromonas hydrophila*

Roslianah Asdari and Mohammad Bodrul Munir

SESSION 5: TRENDS IN FISH HEALTH MANAGEMENT

Diets influencing hematological profile as fitness and genetic bioindicator of fish health

Hon Jung Liew, Okwuosa Obinna Ben and Eyo Joseph E.

Assessment of antibacterial activity of fresh garlic juice extract against *Vibrio spp.* isolated from hybrid grouper (*Epinephelus fuscoguttatus* x *Epinephelus lanceolatus*): an in vitro study

Izzuan-Razali M., Nik-Haiha N.Y., Idris S. M., Azila A., Rimatulhana R., Nur-Nazifah M., Syafiq M. R. M., Sufian M. and Firdaus-Nawi

Comparative study of two host associated probiotics and commercial probiotic effects on growth performance, biochemical composition, gut and muscle histomorphology, and protection against pathogenic bacteria in Malaysian Mahseer, *Tor tambroides* early juveniles

Mohammad Kamruzzaman Hossain, Sairatul Dahlianis Ishak, Shumpei Ichata, Noor Diyana Mat Noordin, Md. Abdul Kader, Mohammad Asmat Ullah, Sharifah Noor Emilia, Yeong Yik Sung and Ambok Bolong Abol-Munafi

Naturally occurring and removal of a cranial papilloma of an aquarium held giant grouper, *Epinephelus fuscoguttatus* (Forsskål, 1775)

Kua Beng Chu, Rohaiza Asmini Y, Nur Ashikin A, Marjorie C & Mohd. Zukri Y.

SESSION 6: TRENDS IN SHRIMP HEALTH MANAGEMENT

Lactococcus garvieae: what we know now and what remains to be researched in giant freshwater prawn *Macrobrachium rosenbergii* (de Man 1897)

Omkar Vijay Byadgi, Rubicely Balan, Sudarshan Pandey, Pei-Chi Wang and Shih-Chu Chen

Noni, *Morinda citrifolia* fruit extract as potential antibacterial and increasing resistance of *Penaeus vannamei* postlarvae against *Vibrio parahaemolyticus*

Julia Hwei Zhong Moh, Yeong Yik Sung and Mhd Ikhwanuddin

Sustainable extraction of natural immunostimulants from shrimp waste

Hajar Rastegari, Su Shiung Lam and Meisam Tabatabaei

Sustainable shrimp management: the critical role of advanced sustainability assessment tools

Meisam Tabatabaei

***Vibrio* species identified as possible component causes of shrimp white feces syndrome associated with the microsporidian *Enterocytozoon hepatopenaei* (EHP-WFS)**

Natthinee Munkongwongsiri, Naing Win Htut, Rungkarn Suebsing, Kanokwan Lertsiri, Kallaya Sritunyalucksana, Anuphap Prachumwat

Effect of oral administration of *Zingiber officinale* extract on growth and immune functions of shrimp *Penaeus monodon* against white spot syndrome virus (WSSV)

Alokesh Kumar Ghosh, Shaikh Shaon Ahmmed, H M Rakibul Islam, Ghausiatur Reza Banu, Sujogya Kumar Panda, Walter Luyten

ePoster: Biosecurity in Aquaculture

Economic impact of *Vibrio* vaccination on cage-cultured of Asian seabass (*Lates calcarifer*)

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In Malaysia, Asian seabass (*Lates calcarifer*) is widely farmed and became the most preference species in the fish market. However, Asian seabass are prone to diseases mainly vibriosis. Vibriosis outbreak may cause additional cost, tremendous mortality and huge economic losses to the farmers. This study aims to estimate cost of *Vibrio* vaccination in grow-out of Asian seabass (*Lates calcarifer*) cage-cultured farm by using stochastic bioeconomic model. *Vibrio* vaccination were given by using oral method on week 0, 2 and 6 of the sampling periods. Furthermore, the prevalence of *Vibrio* spp. between vaccinated and non-vaccinated groups are identified from fish intestines through metagenomics approaches with the present of potential biomarkers. Then, the results were added in the stochastic bioeconomic model developed in Microsoft Excel® (Microsoft Corp., Redmond, WA, USA) using @Risk (Palisade Corp., Ithaca, NY, USA) add-on. Model inputs were based on previous literatures, information from farmers on the farm and expert opinions. Model built comprised biological parameters such as prevalence of vibriosis, mortality and body weight variations, therefore provides acuity in costs of vibriosis and feed. Output from the stochastic bioeconomic model is analyze descriptively using @StatTools add-on (Palisade Corp. Ithaca, NY, USA) in Microsoft Excel® (Microsoft Corp. Redmond, WA, USA.) where cost of rearing vaccinated fish is calculated. Simulated bioeconomic model showed 71.01% of Asian seabass survived meanwhile mortality rate of Asian seabass due to vibriosis and other reasons are 16.60% and 12.40% respectively. Cost of vibriosis and other reasons are the cost to be incurred by farmer's equivalent to RM 0.70/tail/kilogram and RM 0.36/tail/kilogram, respectively. The cost of variables, fixed costs, and provision costs for Asian seabass cage-cultured operations are calculated as RM 4.68, RM 8.79, and RM 1.06 per tail, respectively. Thus, vibriosis vaccination should be applied on a farm with outbreaks to obtain precise result on the efficacy of vaccine as well to ensure the estimated cost per usage are worth for overall production cost.

Keywords: Asian seabass; *Lates calcarifer*; vibriosis; vaccine; stochastic bioeconomic model

Short Biography

Arbania is currently pursuing her Masters at the Universiti Putra Malaysia. She received her Bachelor's degree on 2019 from the Universiti Malaysia Terengganu in Applied Science of Fisheries.

Biofloc composition in whiteleg shrimp, *Penaeus vannamei* culture system as biosecurity approach towards sustainable shrimp aquaculture production

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A study on the biofloc composition from whiteleg shrimp, *Penaeus vannamei* culture pond located in Yan, Kedah was conducted in order to identify the biofloc function as the biosecurity towards prevention of disease in the shrimp culture pond. Sample of biofloc was collected from Pond 2B during DOC 30, DOC 60 and DOC 90 which further identified using Advance microscope *Nikon 80i*. From the study, the biofloc composition consists of microalgae from Bacillariophyte group such as *Leptocylindrus* sp, *Cymbella* sp. *Cocinodiscus* sp, *Thalassiosira* sp., *Nitzschia* sp. and *Amphora* sp. The green algae, Chlorophyte group from *Chlorella* sp. and Cyanobacteria, Cyanophyte group from species *Gleocapsa* sp., *Oscillatoria* sp. and *Anabena* sp. were also identified in the biofloc samples. The zooplankton such as rotifer and protozoa from Ciliophora group, *Vorticella* sp. were also identified in the biofloc samples. The marine worm from group Chaetognatha and nematode also identified in the samples. Overall, it can be concluded that the biofloc composition contains variety and diversity of phytoplankton and zooplankton as the natural diet to the shrimp, even though contains other type of protozoa such as the vorticella and nematode in the samples. The appearance of the protozoa and nematode identified as the algae grazer due to the dense of microalgae and phytoplankton aggregated in the biofloc. Hence, it can be concluded that the biofloc system assist in sustaining a good biosecurity for disease prevention in the shrimp aquaculture system, while maintaining a good water quality and help in sustaining a good performance shrimp in the culture ponds.

Keywords: Phytoplankton, *P. vannamei*, zooplankton, protozoa, biosecurity, sustainability

Short Biography

Dr. Hidayah Manan is currently working as Postdoctoral at the Institute of Tropical Aquaculture and Fisheries (AKUATROP), Universiti Malaysia Terengganu. She received her Doctoral degree, PhD in Sc. Aquaculture from University of Malaysia Terengganu. Dr Hidayah also completed her Masters in Sc. Aquaculture from UMT. Dr. Hidayah has authored numbers of publications in various journals and books. Her publications reflect her research interests in Biofloc technology, Green Technology in Aquaculture, and also in the Marine shrimp culture system.

Fatty acid compositions and oil quality assessment of liver of yellow puffer, *Xenopterus naritus*

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Yellow puffer fish, *Xenopterus naritus* are among common fish caught at coastal waters of Kuching and Samarahan District. The aims of the present study to determine fatty acid composition and to evaluate oil quality indices of puffer liver. Two methods were used for oil extraction, namely as boiling procedure and Bligh & Dyer cooking-pressing method. For each 50 g liver, approximately 10.13% and 21.51% of total yield fish oil were extracted in each respective method. Purified oil extraction were subject to fatty acid analysis using GC/MS. The result from GC/MS analysis has shown the specific fatty acid composition of puffer liver oil and their enrichment in MUFAs and PUFAs, with special reference to omega 3 and 6. Based on oil quality indices, the acid value, free fatty acid, peroxide value and saponification value were within acceptable range. This preliminary study suggests the potential use of puffer liver as fish oil source.

Keywords: Puffer fish, fatty acid, cooking-pressing, GC/MS, oil quality indice

Short Biography

Dr. Samsur Mohamad is currently working as Senior Lecturer at the Universiti Malaysia Sarawak. He received his Doctoral degree or PhD on Fisheries Science from the University of Nagasaki Japan. Dr Samsur Mohamad completed his Masters Fisheries Science from the University of Nagasaki, Japan. He then worked at the Universiti Malaysia Sarawak, served as Senior Lecturer at the Faculty of Resource Science and Technology. Dr. Samsur Mohamad has authored several publications in various journals and books. His publications reflect his research interests in Marine Biotoxins and Fisheries Sciences.

Advances in technologies towards enhancing shellfish wellbeing for optimum aquaculture production

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Aquaculture production has been in existence since 12th century, though unpopular due to the flourishing of capture fisheries. Up to 2000's, debates on the potentials of aquaculture as an alternative to artisanal fisheries has been intense. When *Nature* journal published a review on the sustainability and contribution of aquaculture to the world from 1997 to 2017, many countries of the world became attracted to the sub-sector, which saw its rapid development. The global aquaculture development has never been without hurdles emanating from its operations. Challenges such as proper nutritious feed, development of antibiotics and vaccines to fight diseases and parasites, sustainable breeding techniques, social conflicts, environmental impacts, availability of good water and culture facilities as well as economic and marketing issues have kept researchers on their feet over the years in attempt to find lasting solutions. Consequently, numerous technological advancements have been recorded in recent years. Introduction of gene technology, Internet of Things (IoT), transcriptomes, nanotechnology, mechanical filters and biological filters in Recirculation Aquaculture System (RAS), Supercritical Fluid (SCF) extraction process and high-throughput messenger RNA (mRNA) sequencing (RNA-seq) facilities among others have indeed broaden the prospects of an eco-friendlier and more sustainable aquaculture to meet the Food and Agriculture Organization (FAO) goal of 2050.

Keywords: Advance aquaculture, gene technology, Internet of Things (IoT) and Recirculation Aquaculture System (RAS)

Short Biography

Mr Benedict Iber is a lecturer with Federal University of Agriculture Makurdi, Benue State, Nigeria. He is a PhD student at the prestigious institute of Tropical Aquaculture, Universiti Malaysia Terengganu. Mr Benedict is currently working on the chemical isolation and characterization of natural chitosan from shrimp and prawn for aquaculture wastewater treatment and subsequent application of recovered solids in crop production and soil conditioning. With vast knowledge in Fisheries and Aquaculture from his first and masters degrees, he has researched and published in many areas of Aquaculture including nutrition, limnology, fish biology and Biotechnology.

Sustainable fishery and aquaculture development of yellow pufferfish from Sarawak, Malaysian Borneo

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Yellow pufferfish, *Chonerhinos naritus* (Richardson, 1848) can only be found exclusively in the state of Sarawak, Malaysian Borneo. This species is important in subsistence fisheries and commonly inhabit the shallow coastal waters and moves further upstream into Saribas River for spawning. *C. naritus* is a commercially important species in subsistence fisheries because it has been consumed by the locals for many generations. Despite the presence of Tetrodotoxin (TTX) existed in the species, no poisoning case was reported due to the proper preparation of the fish, making it suitable to be introduced as a new candidate in aquaculture. Therefore, the main objectives of this study were to determine the population status, investigate the reproductive biology and develop the seed production technology of *C. naritus*. Sarawak coastal waters were considered to be favourable to the species based on the positive allometric growth in length-weight relationship analysis (LWR) and condition factor (CF). *C. naritus* appeared to be a species with sexual dimorphism in size and showed five stages each for ovarian and testicular development, namely immature, developing, spawning capable, regressing and regenerating stages. Gonadosomatic index (GSI) showed the reproductive period occurred between July and August every year. The propagation of *C. naritus* followed the same general pattern in other farmed fish species, except that the sexually mature broodstock were collected immediately prior to spawning for gamete collection, thus not required any hormone administration. Results in larval rearing experiment indicated the optimal photoperiod was in total darkness (0h light:24 h dark), temperature from 26 to 28 °C and salinity ranges from 15 to 20 ppt. In conclusion, this study clearly indicates that albeit there were many further works needs to be done, *C. naritus* remains a very attractive candidate for aquaculture. Comprehensive study and consistent efforts to improve the existing culture protocols are crucial for further development of *C. naritus* aquaculture.

Keywords: Yellow pufferfish, *Chonerhinos naritus*, aquaculture, Sarawak, reproductive biology.

Short Biography

Dr. Ahmad Syafiq is a lecturer at Universiti Malaysia Sarawak (UNIMAS). He received his Doctoral degree in Aquaculture from the same university after completing his MSc in aquaculture from National Taiwan Ocean University. He has worked as a technical executive at Sea Party Technology R&D Sdn. Bhd. in Tanjung Manis Halal Hub, under Sarawak Corridor of Renewable Energy (SCORE). Dr. Ahmad Syafiq has authored several publications in various journals and books. His publications reflect his research interests in fish reproductive biology, finfish breeding techniques and community-based sustainable aquaculture

Gamma ray irradiation: A valuable tool for fresh feed disinfection

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Shrimps aquaculture remain one of the important sector in the aquaculture industry in Malaysia as of 2015 to 2018 Malaysia produced 187,404.97MT with the value of RM4,238,315,430.00 as according to annual fisheries statistic, Department of Fisheries, Malaysia. However, most of the hatchery operators rely on domesticated SPF shrimp broodstock imported from broodstock multiplication center (BMC) outside of Malaysia. These broodstocks are expensive ranging from USD100 to USD200 per piece. Hatchery operators spend more than RM 100,000.00 on a single shipment. Despite being SPF, somehow, outbreak of diseases such as, early mortality syndrome (EMS), whites spot (WSSV) and *enterocytozoon hepatopenaei* microsporidia (EHP) infection occurred at farm level. It could be due to management issue at farms. However, the hygiene of fresh feed at hatcheries it is often overlooked, this fresh feed has been documented to be a potential source of host for pathogens particularly for marine polychaetes and squids. There is no procedure to disinfect prior to use at private hatcheries. At FRI Pulau Sayak, these fresh feeds were disinfected by gamma irradiation prior to feeding. Gamma ray irradiation on positive sample proved to be effective to disinfect potential pathogens. PCR screening on fresh feed showed that gamma ray irradiation successfully disinfected pathogen in the fresh feed. Gamma radiation destroy the pathogen's DNA directly or by the effect of water ionization that produce free radicals. The effective dose is between 5 – 10 kGy of gamma ray from cobalt (⁶⁰Co). Gamma ray irradiation is new method for food disinfection in Malaysia, nevertheless it is crucial for the local aquaculture industry to ensure hygienic feed for the broodstocks be it shrimps or fishes.

Keywords: Gamma ray irradiation, domesticated SPF shrimp broodstock, fresh feed, *enterocytozoon hepatopenaei*, early mortality syndrome, White spot syndrome virus,

Short Biography

Teoh Pik Neng is currently working as research officer at the Fisheries Research Institute, Department of Fisheries, Malaysia. He received his degree in Aquatic Biology from the University of Science, Malaysia. Teoh Pik Neng has been working on a broodstock development programme on shrimps and research in microalgae production.

Evaluating antibiotic susceptibility profile of *Vibrio* species isolated from cultured *Epinephelus* species in Peninsular Malaysia

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Farmers regularly employ antibiotics because they can be used for both preventative and therapeutic purposes in the fight against bacterial growth. Antibiotic overuse in the aquatic environment and aquaculture has resulted in the establishment of antimicrobial-resistant (AMR) bacteria. Thus, it is essential to investigate the resistance level of *Vibrio* spp. against antibiotics to provide a current situation on antimicrobial resistance strains in Malaysian farms. Enhancement of grouper farming due to high human food demand requires a comprehensive database on the antibiotic-resistant level of *Vibrio* spp. for appropriate antibiotic administration in disease treatment. In this work, *Vibrio* species were isolated using 270 cultured groupers taken from nine farms in Peninsular Malaysia. Analysis of the antimicrobial resistance profile suggested that ampicillin and penicillin G were ineffective in treating *Vibrio* infections since a high number (80%) of *Vibrio* spp. were resistant to both antibiotics. Effective monitoring on the administration of the bacitracin, erythromycin and vancomycin was required since 30% to 54% of the *Vibrio* isolates were resistant to these antibiotics. Meanwhile, *Vibrio* spp. resistance to tetracycline and streptomycin remained modest (14 %). The majority of the species tested were very susceptible to both antibiotics, including *V. parahaemolyticus*, *V. alginolyticus*, *V. rotiferianus*, *V. campbellii*, and *V. diabolicus*. According to the multiple antibiotic resistance (MAR) index, 88% of *Vibrio* spp. exhibited a MAR index value of more than 0.2. As a result, the findings suggested that a large number of *Vibrio* spp. were resistant to several antibiotics and had been exposed to antibiotics on a regular basis at the farms. Plasmid profiling revealed that 61% of *Vibrio* spp. were chromosomal-mediated (MAR index 0.37), while 39% were plasmid-mediated (MAR index 0.56). Most *Vibrio*-positive plasmids lost their antibiotic resistance after the curing process, and the AMR index was lowered to 0.21. As a result of the findings, it appears that the presence of plasmids conveying antibiotic resistance can increase *Vibrio* spp. antibiotic resistance. Awareness of the problem of antibiotic resistance among farmers, as well as ongoing monitoring of antibiotic-resistant microorganisms, particularly on specific antibiotics, are useful for rules governing farm practices.

Keywords: Vibriosis, Grouper *Epinephelus* sp., antibiotics, antimicrobial-resistant (AMR) bacteria, multiple antibiotic resistance (MAR) index

Short Biography

Dr Ina-Salwany is currently working as a Senior Lecturer and the Head of Aquatic Animal Health and Therapeutics Laboratory at Universiti Putra Malaysia (UPM). She received her Doctoral degree from Universiti Malaysia Terengganu in Molecular Biology. She has published numerous publications in various national and international peer-reviewed journals and presented scientific papers across the world. Her publications reflect her research interests in Fish Molecular Biology.

Parasite Infestation in *Macrobrachium rosenbergii* from Sarawak (Kuching-Samarahan Region)

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Macrobrachium rosenbergii has proven to be one of the important species in Malaysia, since it is a significant contributor to Malaysia's aquaculture sector, and as the source of food for the Sarawak people. However, due to parasite infestation, this species has been threatened with low productivity and quality. Hence, present study focuses on the parasites infesting the *M. rosenbergii* and the relationship with the body size of the prawns obtained from Sarawak, precisely from the Kuching-Samarahan region. A total of 72 samples were dissected and processed in the lab to analyse their body length, weight and for the presence of parasites. Results found that eleven individuals were infesting about 15% (11 out of 72) of the samples which were identified as *Probopyrus* sp., *Calanus* sp., Nematode, and an unknown larva. The highest prevalence among these parasites is from *Probopyrus* sp. (6.94%), followed by Nematode (4.17%), *Calanus* sp. (2.78%), and unknown larva (1.39%). Majority of the infected prawns were categorised under the 111 - 120 mm interval, which is the shortest length group with an average weight of 17.91 g and none were infected from the bigger prawns. The correlations between the parasite mean intensity and the prawn length (mm) were found to be negative for all of the parasites mentioned; *Probopyrus* sp. ($r = -0.725$), *Calanus* sp. ($r = -0.518$), nematode ($r = -0.646$) and the unknown larva ($r = -0.137$). The study of *M. rosenbergii* from Sarawak requires more research to better understand its correlation with parasite infestation by determining the impact of the present water quality index of the researched areas on parasite prevalence.

Keywords: *Macrobrachium rosenbergii*, Parasites, Prevalence, *Probopyrus* sp., *Calanus* sp

Short Biography

Rizmanisha Jasmin Ruzaiuddin is currently a final year student at the University of Malaysia, Sarawak, completing her Bachelor of Science with Honours in the Aquatic Science and Resource Management Programme. Her final year project report reflects her interest in studying parasites.

Shrimp with positive PCR for DIV1-ATPase gene revealed no pathological lesions of DIV1

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Decapod iridescent virus 1 (DIV1) is an emerging virus that has caused massive mortality in many economic crustacean species in China including *Cherax quadricarinatus*, *Macrobrachium nipponense*, *M. rosenbergii*, *Procambarus clarkia*, *Exopalaemon carinicauda*, *Penaeus vannamei* and *P. monodon*. The infected animals demonstrate an empty stomach and midgut accompanied with a pale hepatopancreas and soft shell. The World Organization for Animal Health (OIE) recommends a DIV1-PCR detection method targeting the DIV1 ATPase gene. Here we describe farmed shrimp samples of *P. vannamei* that were collected weekly and tested for DIV1 ATPase gene by PCR during cultivation over two crop cycles in Thailand. Some specimens (20-60%) from both crops gave positive PCR test results for DIV1 ATPase during cultivation. However, histological analysis revealed no characteristic DIV1 lesions in their hematopoietic tissue (HPT) or lymphoid organs (LO). Absence of DIV1 was also confirmed by negative ISH results in the HPT and LO. In addition, there was no significant mortality observed in both crop cycles. Altogether these results indicate that false positive results can be obtained from healthy shrimp. It is recommended to be careful in using only one PCR protocol to detect DIV1 infection in shrimp.

Keywords: Decapod iridescent virus 1 (DIV1); non-infectious; shrimp; *Penaeus vannamei*

Short Biography

Dr. Suebsing received her D.Sc. degree in Marine Biotechnology from Gangneung-Wonju National University, Korea in 2012. From 2012 to 2014, Dr. Suebsing's research is focused on biosensors for aquatic animal pathogens and translational research based on shrimp-pathogen interaction. Recently, she is a researcher at National Center for Genetic Engineering and Biotechnology (BIOTEC), National Science and Technology Development Agency (NSTDA), Thailand.

First report of Sacculinidae parasite infestation on crab population (Subfamily Thalamitinae) in Peninsular Malaysia.

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Sacculinidae is one of the parasitic group family lies under Rhizocephalan which is a threat to the wild population of the crabs due to the ability to hold the development of the gonad in female crab and also can cause the abnormal shape of the male crab abdomen (wider and globular like the female crab abdomen) in the previous study. The previous report mentioned that there is another infestation of *Sacculina beauforti* on mud crab, *Scylla olivacea* (Subfamily Portuninae), occurring in Sabah. The infestation of this parasite can cause a decrease in the crab population in the natural habitat due to the inability to mating. Screening of the crab under family Thalamitinae was conducted in two locations (n=300 for each sample size) Malaccan Strait and the South China Sea representing Peninsular Malaysia. Out of 600 specimens, only 38 crabs (6.3%) (20 males and 18 females of the same species) were found to be infected by Sacculinidae parasite. The samples were collected and subjected to DNA extraction using commercial kits. The total genomic DNA was amplified using COI universal primer and sent for sequencing. The COI DNA sequence was subjected to phylogenetic construction and showed a monophyletic clade with other Sacculinidae indicating that this parasite belongs to the genus *Sacculina*. We are still working out to characterize the species of this parasite and the prevalence of this parasite according to seasonal changes.

Keywords: *Sacculina sp.*, Thalamitinae, Infestation, crab, Peninsular Malaysia

Short Biography

Mr Fazhan Hanafiah is currently working at the Institute of Tropical Aquaculture and Fisheries University Malaysia Terengganu (UMT). He received his PhD from UMT in 2016. His research focus on Crustacean Aquaculture and Genetic breeding, and also marine biology field. His publications mostly focus on the Mud crab genus *Scylla* and other brachyuran crab as well.

ePoster: Detection Method/Diagnostic (Parasitic, Bacterial & Viral Diseases)

The externa histological description of a sacculinid (Crustacea; Rhizocephala) and its impact on the gonadal development of mud crab

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Rhizocephalan parasites are known to infect other crustaceans and often result in serious morphological, physiological, and behavioral changes upon infection. The most notable feature of infected crabs is the development of an externa (female reproductive organ) on the outer layer of the host's abdomen. This study provides the first detailed description of the externa of *Sacculina beauforti* on the orange mud crab *Scylla olivacea*. In addition, the gonadal tissues of infected and non-infected individuals were also compared. We noticed that rootlets were noticeable in the testicular cross-sectioning, whereas female reproductive organs degenerated upon infection. The results of this study contribute toward the taxonomic characterisation of the sacculinid rhizocephalan and the disruption of *S. beauforti* towards the reproductive biology of its host.

Keywords: sacculinid, Rhizocephala, mud crab, externa histological

Short Biography

Dr. Khor Wai Ho is a researcher/senior lecturer at the Institute of Tropical Aquaculture and Fisheries, Universiti Malaysia Terengganu. He graduated from UMT with a PhD in Aquaculture, and completed a two-year postdoctoral at Shantou University, China in the field of marine biology. His main research focus is on crustacean biology and physiology, biotechnology and aquaculture. He has published more than 60 WoS-indexed papers and is currently the associate editor for several high impact journals, including Biodiversity and Conservation, Frontiers in Marine Science, and PeerJ.

Microbiome analysis of gut bacterial communities of healthy and diseased Malaysian mahseer (*Tor tambroides*) using 16S rRNA metagenomics approach

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The gut microbiota is referred to an 'extra organ' and is critical in assisting the host in terms of nutrition and immunity. Environmental stressors could alter gut microbial community and cause gut inflammation. This study aimed to investigate and compare the gut microbiota community between healthy and diseased *Tor tambroides*. In this study, such gut microbial alterations were explored using NGS-based 16S rDNA sequencing on the Malaysian mahseer (*T. tambroides*). Three adult healthy and three diseased adult Malaysian mahseers (showing signs of exophthalmia, coelomic distension and petechial haemorrhage) were obtained from LTT Aquaculture Sdn Bhd. Our results revealed significant differences in microbial diversity, composition and function between both populations of *T. tambroides*. Alpha diversity analysis depicts lower diversity of gut microbiota composition in diseased *T. tambroides* as compared to the healthy group. In particular, Enterobacteriaceae, Aeromonas, Bacteroides, Vibrio and Pseudomonas were found within gut microbiota of the diseased fishes. In addition, cellulose-degrading bacteria and protease-producing bacteria were identified from the gut of *T. tambroides*. Thus, our findings emphasised on the association between the alteration in gut microbiota composition and infectious abdominal dropsy (IAD) in *T. tambroides*. This finding is important to provide basic information for further diagnosis, prevention and treatment of intestinal diseases in fish.

Keywords: 16S rRNA gene, gut microbiota, infectious abdominal dropsy, Malaysian mahseer, metagenome

Short Biography

Dr. Scientist is currently working as a senior lecturer in the Universiti Malaysia Sarawak at the Faculty of Resource Science and Technology. He received his Doctoral degree or PhD on Healthcare Biotechnology from the Universiti Sains Malaysia. He then worked at the Lembaga Getah Malaysia serving as a research officer at the Biotechnology Unit. Dr. Chung has authored several publications in various journals and book on freshwater fishes in Borneo. His publications reflect his research interests in using molecular biology to decipher the uniqueness of freshwater fishes in Borneo.

Pathogenicity of different betanodavirus strains in Asian Seabass (*Lates Calcarifer*) under temperature fluctuation stress

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Viral Nervous Necrosis (VNN) disease that is caused by betanodavirus infection are responsible for mass mortality in many marine cultured fish species. . Fish can acquire the diseases by either horizontal or vertical transmission. Malaysian local betanodavirus strains were obtained from National Fish Health Research Centre, Penang and cultured in E-11 cell line. The isolates are Bawal emas (BE), AVA and KL 22. Fifty-percent tissue culture infective dose (TCID₅₀) calculations for all the isolates was conducted to evaluate the concentration of the virus. In this study, three hundred and sixty healthy seabass juvenile (5 ± 2 g) with size two to three inches were obtained from Marine Fish Aquaculture Research Division, Terengganu were divided equally in eight tanks that represent four treatment groups with duplicates; Group 1 (AVA strains), Group 2 (BE strains), Group 3 (KL22 strains) and Group 4 (control). All the fishes were observed daily for any clinical changes and mortality occurred. Upon clinical sign appear and mortality, the fish were slaughtered immediately and subjected to polymerase chain reaction (PCR), cell culture and histology. Experiment was terminated after thirty days period. TCID 50 for all the tested strains are as follow; KL 22 = 10^4 TCID₅₀/mL, BE = 10^7 TCID₅₀/mL and AVA = 10^6 TCID₅₀/mL. KL22 strain was found to have the highest TCID 50 followed by AVA and BE. Similarly, the highest mortality also recorded in the group challenged with KL22 strain that also has the highest TCLD 50. All the dead and sick fishes were tested positive for PCR. However, the histology finding indicated no vacuolation found in the cell suggested an acute infection was occurred.

Keywords: VNN, betanodavirus, Asian seabass, TCID50

Short Biography

Safwan is currently working as research assistant at the International Islamic University Malaysia.

Improvement of a method for detecting *Perkinsus olseni* in low infection intensity samples

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Parasite *Perkinsus olseni* has listed as a pathogen notifiable under aquatic animal health in World Organization for Animal Health (OIE). Microscopic detection of prezoosporangia using Ray's fluid thioglycollate medium (RFTM) is recommended as the most sensitive genus-specific detection method. However, the genus-specific PCR assay is not sensitive enough to detect low infection intensity samples. In this study, the detection and identification method of *P. olseni* was improved for low infection intensity samples by combining prezoosporangia production by RFTM and the species identification by sequencing of the genus-specific PCR products from prezoosporangia. A wide range of numbers of prezoosporangia were subjected to DNA extraction and the ITS region of *Perkinsus* was amplified with the genus-specific PCR. Results shown the detection of *P. olseni* were achieved with the minimum amounts of 3, 5 and 10 prezoosporangia with KOD One, GFlex, and MyTaq, respectively. The detection method of *P. olseni* was established in this study by combining RFTM assay, the genus-specific PCR on prezoosporangia, and the sequencing of PCR products, even in low infection intensity samples of 3 prezoosporangia. The improvement method can be used in surveillance program of *P. olseni* in molluscs.

Keywords: *Perkinsus olseni*, Ray's fluid thioglycolate medium (RFTM), ITS region, low infection intensity samples, prezoosporangia

Short Biography

Vui Kien Liew is currently working as Fishery Officer at the Department of Fisheries, Malaysia. He completed his Masters of Agricultural and Life Sciences from the University of Tokyo, Japan.

ePoster: Prevention & Control Measures (Parasitic, Bacterial & Viral Diseases)

Production of biochar via microwave pyrolysis of palm kernel shell for simultaneous removal of ammonia and cultivation of lettuce in aquaponic system

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A recirculating aquaponic system comprising of ammonia excretions from farming of African catfish was developed in this study to convert bacterial into nitrates, followed by the use of nitrates as nutritious ingredient to nurture lettuce via hydroponic approach. Microwave pyrolysis of palm kernel shell combined with physical activation was performed to produce microporous biochar (1.803 nm of pore width) with high BET surface area up to 419 m²/g). The biochar was found desirable as biological carrier to cultivate nitrifying bacteria which improved the quality of aquaculture wastewater by eliminating total suspended solids (68%) and ammonia (67%). As a result, low amount of total suspended solid (59.40 mg/L) and ammonia (0.42 mg/L) were found in the African catfish wastewater. The biochar also transformed the ammonia into nitrate (29.7 mg/L), and enhanced the nitrogen uptake by the lettuce (110 mg of nitrogen per plant). This has led to high production of lettuce (0.0562 %/day), whilst sustaining acceptable level of BOD₅ (3.94 mg/L) and 100% of catfish survival rate. This study revealed that biochar produced from microwave pyrolysis of palm kernel shell can be applied in aquaponic system as nitrifying bacteria growth medium for removal of ammonia in aquaculture wastewater, while generating nitrate for cultivation of vegetables that support food security.

Short Biography

Wan Adibah Wan Mahari is currently a post-doctoral at the Institute of Tropical Aquaculture and Fisheries (AKUATROP), Universiti Malaysia Terengganu. Wan Adibah has received international and local awards and honours in academia, research and innovation such as Vice Chancellor Champion Trophy, Grand Prize Award, Gold Award, Silver Award, Bronze Award. In 2022, she is selected as participant by the National Research Foundation, Prime Minister Office of Singapore to engage with eminent scientific speakers, include recipients of the Nobel Prize, Fields Medal, Millennium Technology Prize and Turing Award in Global Young Scientist Summit 2022. Her success spurred her interest to delve into research matters in more detail. Through her ongoing research on developing an integrated microalgae-shellfish cultivation system, Wan Adibah is paving the way for advancements in shellfish green technology.

Immune response and tolerance of white-leg shrimp *Penaeus vannamei* to *Vibrio parahaemolyticus* following exposure to *Pandanus tectorius* leaf extract

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The effect of *Pandanus tectorius* leaf extract on the White-leg shrimp *Penaeus vannamei* tolerance against *Vibrio parahaemolyticus*, a causative pathogen for vibriosis, were investigated in this study. Specifically, thirty shrimp post-larvae (1 cm) were exposed to 0.5, 1, 2, 3, 4, 5 and 6 g/L leaf extract for 24 h and subsequently observed for survival and immune-related genes expression (Hsp70, ProPO, peroxinectin, penaeidin, crustin and transglutaminase), followed by determination of their tolerance and histological tissue profiles upon *Vibrio* challenge. Survival of shrimps treated with 6 g/L of leaf extract, the highest dose examined in this study, improved by up to 95% to controls, which were not exposed to the leaf extract. Hsp70, crustin, and prophenoloxidase mRNA levels were observed to be 8.5, 10.4, and 1.5-fold higher, respectively, when immune-related genes were quantified by qPCR. Histopathological analysis of the hepatopancreas and the muscle tissues revealed major tissue degeneration in *Vibrio*-challenged shrimps but not in shrimps primed with *Pandanus tectorius* leaf extract. Of all the dose examined, the best pathogen resistance results were obtained with a 24 h incubation of shrimp in 6 g/L *P. tectorius* methanolic leaf extract. The tolerance towards *V. parahaemolyticus* might be associated with the increased regulation of Hsp70, prophenoloxidase and crustin upon exposure to the extract, all immune-related proteins essential for pathogen elimination in Penaeid shrimp.

Keywords: *Penaeus vannamei*; *Pandanus tectorius*; *Vibrio*; Immunity; Hsp70

Effect of dietary prebiotics on snakehead (*Channa striata*) health: haematology and disease resistance parameters against *Aeromonas hydrophila*

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This study examined the effect of dietary prebiotics after 16 weeks, followed by 8-week post-feeding trial along with a control (non-supplemented) with haematology study for disease resistance and immune response in *Channa striata* fingerlings against *Aeromonas hydrophila*. Fish were raised on different feed (40% protein and 12% lipid) which incorporated three different prebiotics (β -glucan, galacto-oligosaccharide and mannan-oligosaccharide) and a control. Supplementation with dietary prebiotics led to improving significantly ($P < 0.05$) the red blood cells, white blood cells, packed cell volume, haemoglobin concentration and serum protein level and lysozyme activities. These improvements were especially effective ($P < 0.05$) when the fish were challenged with *A. hydrophila* at the dose of 2×10^6 . The disease resistance against *A. hydrophila* was significantly ($P < 0.05$) higher in fish fed with prebiotic supplement (β -glucan) compared to other two prebiotics and the control. The study showed no differences in the efficacy of β -glucan, GOS and MOS after post-feeding with an unsupplemented feed over a longer time duration, which was first reported for this species.

Keywords: Asian snakehead, *Channa striata*, dietary prebiotics, haematology

Short Biography

Dr. Roslianah Asdari is currently working as a lecturer in the Faculty of Resource Science and Technology at Universiti Malaysia Sarawak, Sarawak, Malaysia. Dr. Roslianah completed her Masters on Aquatic Biology from the Universiti Sains Malaysia, Pulau Pinang, Malaysia. She received her Doctoral degree or PhD on Agricultural Science (Fisheries Science) from Kindai University, Osaka, Japan in 2014. She then worked at UNIMAS as a lecturer since 2016 until now. Dr. Roslianah has authored several publications in various journals. Her publications reflect her research interests in aquaculture fish nutrition and feeding management.

ePoster: Trends In Fish Health Management

Diets influencing hematological profile as fitness and genetic bioindicator of fish health

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Hematology analysis is one of the validated technique use in fish health evaluation and welfare in veterinary practice for; aquaculture and scientific research. Studies have shown that hematological parameters have proven to be highly relevance to various environmental factors including water quality, nutrition, stress or pathogens etc. Blood analysis provide essential information about the physiological aspects of fish health indication such as the activation status of the neuroendocrine and immune system, acute and long-term stress impacts due to adverse husbandry conditions, potential diseases infection and genetic predispositions. Hematological indices include leukocytes (white blood cells (lymphocytes specifically)) for immunity, thrombocytes (platelets) for blood clotting, erythrocytes (red blood cells) for oxygen circulation, neutrophils, monocytes and packed cell volume. Other calculable indices including mean corpuscular volume, mean corpuscular haemoglobin concentration and mean corpuscular haemoglobin. The aim of this review was to summarize the hematological profile of fish as physiological and genetic bioindicator in fish influence by diets and health status. We therefore recommend that more attention should be paid to the relationship between fish haematological indices, diets and influence of genetic make-up on welfare and health. We believe that scientific investigation on the effect of fish diets on hematological profiles in fish health and molecular assessment should be encouraged.

Key words: Hematology, fish health, fish physiology, gene expression

Assessment of antibacterial activity of fresh garlic juice extract against *Vibrio* spp. isolated from hybrid grouper (*Epinephelus fuscoguttatus* x *Epinephelus lanceolatus*): An *in vitro* study

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We assessed the antibacterial activity of fresh garlic juice extract against *Vibrio* spp. isolated from hybrid grouper (*Epinephelus fuscoguttatus* x *Epinephelus lanceolatus*). The disc diffusion method was conducted to evaluate five concentrations of fresh garlic juice extract against *Vibrio alginolyticus*, *Vibrio vulnificus*, and *Vibrio parahaemolyticus*. The tested concentrations are 20%, 40%, 60%, 80%, 100%, and oxytetracycline (OTC) antibiotic as a positive control, while absolute ethanol as a negative control. Results from the disc diffusion study revealed 100% and 80% garlic juice extract have higher inhibition zone against *V. alginolyticus* and *V. vulnificus* compared to OTC. The inhibition zone against *V. parahaemolyticus* was slightly less when 100% garlic juice extract gave an equal inhibition zone with OTC and smaller in other concentrations. Minimum inhibition concentration (MIC) and minimum bactericidal concentration (MBC) were also studied, which resulted in the lowest MIC value was *V. alginolyticus* (0.011%) followed by *V. vulnificus* (0.023%) and the highest is *V. parahemolyticus* (0.045%). Similarly, the lowest MBC value was *V. alginolyticus* (0.023%) followed by *V. vulnificus* (0.045%) and the highest is *V. parahemolyticus* (0.09%). The results revealed that fresh garlic juice extract was effective against the test bacteria and showed the potential of using fresh garlic juice extract treatment as an antibacterial agent to treat vibriosis in hybrid grouper fish.

Keywords: Garlic, Hybrid Grouper, *Vibrio* spp., Antimicrobial activity

Short Biography

Graduated with a Bachelor of Marine Science from the International Islamic University of Malaysia. Currently pursuing a master's degree in biotechnology at the same university and working on water quality research at National Fish Health Institute in Penang, Malaysia.

Comparative study of two host associated probiotics and commercial probiotic effects on growth performance, biochemical composition, gut and muscle histomorphology, and protection against pathogenic bacteria in Malaysian Mahseer, *Tor tambroides* early juveniles

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The efficacy of probiotics depends on many factors such as, origin of probiotics, age, and size of fish, duration, dose, and method of application, and culture environment. A feeding experiment were performed to evaluate the comparative effects of host associated probiotics (HAP) with commercially prepared probiotics on survival, growth performances, biochemical composition, gut, and muscle histo-morphology of Malaysian Mahseer, *Tor tambroides* early juveniles (initial weight 2.53±0.02 g). Control fish received basal feed only (T1), whereas two HAPs and a commercial probiotic were added at 1×10⁸ cfug⁻¹ into basal feed as experimental diets (T2: *Aeromonas* sp. A8-29; T3: *Enterococcus faecalis* strain FC11682; T4: commercial probiotics *Bacillus* sp.). After 10 weeks feeding, all treatments were challenged with fish pathogen, *Vibrio parahaemolyticus*, by injecting the amount of lethal dose (LD₅₀:1×10⁸ CFUml⁻¹) intraperitoneally. Results showed that fish fed HAP-supplemented diets performed significantly better in all growth parameters and have higher survival after challenge test. Whole body proximate composition did not differ between treatments. However, both HAP treatments increased total n-6, C18:3n-3, and C18:2n-6 content in whole body, with highest C20:4n-6 content observed in T3 fish. Histology observation showed that both HAP treatments positively affected gut morphology measured by increased villus height, width, area, and also improved hypertrophic muscle growth. In conclusion, the application of HAPs in diets of *T. tambroides* early juveniles, particularly *E. faecalis* strain FC11682, demonstrated overall better performances and could be used as promising probiotics in Malaysian mahseer culture.

Keywords: *Aeromonas* sp., commercial probiotics, disease resistance, *Enterococcus faecalis*; host associated probiotics, *T. tambroides*

Short Biography

Mr. Mohammad Kamruzzaman Hossain (Assistant Director, Department of Fisheries (DoF) Bangladesh) is currently a PhD fellow at the Institute of Tropical Aquaculture and Fisheries (AKUATROP), UMT. He completed his Masters in Aquaculture from Bangladesh Agricultural University. He, then worked at the World FishCenter Bangladesh in controlling WSSV in *Penaeus monodon* farming. Then he joined Bangladesh DoF and start to work as Sub-District level Fisheries Officer. Mr. Kamruzzaman has authored several publications in various journals and books. His publications reflect his research interests in fish health management in Aquaculture.

Naturally occurring and removal of a cranial papilloma of an aquarium held giant grouper, *Epinephelus fuscoguttatus* (Forsskål, 1775)

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A giant grouper, *Epinephelus fuscoguttatus* (Forsskål, 1775) with body weight of 3 kg that held for 4 years in public display aquarium was observed to develop abnormal tissue at the cranial on June 2019. The abnormal tissue was later confirmed as neoplasm. The occurrence of neoplasm was naturally occur and first reported in an aquarium held giant grouper in Malaysia. Physical examination revealed that the papilloma became neoplasm and haemorrhagic after six months later. The cranial neoplasm was approximately 10 cm in diameter, macroscopically, soft and pinkish papillary masses protruded above the normal epithelial surfaces. An attempt to remove the papilloma using ligature was conducted and after 9 days, the neoplasm was disappeared and continued 45 days of observation showed no sign of regrowth of the papilloma.

Keywords: Giant grouper, abnormal tissue, aquarium, cranial papilloma, ligature

Short Biography

Nur Ashikin Arbi is currently working under Parasitology laboratory of National Fish Health Research Division (NaFisH) at Fisheries Research Institute, Department of Fisheries Malaysia. She received her diploma on Civil Engineering from the Polytechnic Kuching Sarawak. She is actively involved in aquatic animal health research focusing on parasite.

***Lactococcus garvieae*: What we know now and what remains to be researched in giant freshwater prawn *Macrobrachium rosenbergii* (de Man 1897)**

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This review on *Lactococcus garvieae* (LG) infection in giant freshwater prawn summarizes current knowledge about the clinical signs, pathogenesis, immune response and immune evasion. It also discusses limitations of existing gaps in knowledge for effective treatment. *Lactococcus garvieae* is an opportunistic pathogen which causes Lactococcosis in *M. rosenbergii*. LG is a catalase-negative, nonhemolytic, cocci shaped bacteria infecting giant fresh water prawn. The wide distribution of *L. garvieae* relates to the ability to adapt and survive in many environmental conditions as well as a wide range of pH (4.5- 9.6), in temperatures of (10°C to 45°C), salinity concentrations (0 to 6.5%). The bacteria cause hyperacute, haemorrhagic septicaemia and leads to high mortality rates. Some of the clinical signs observed during Lactococcosis in giant fresh water prawn are whitish muscle and a swollen, yellowish hepatopancreas, yellowish-white spots on the muscle. Histopathological changes are observed in hepatopancreas and muscles. The hepatopancreatic tubules are severely damaged and haemolytic infiltrations are observed in muscle. The phenoloxidase activity (PO) and glucose level are decreased after infection which shows that *L. garvieae* depresses the immune response and physiological status of giant freshwater prawns. Increase in total haemocyte count and haemolymph protein, Superoxide dismutase (SOD) activity, PO activity are the host immune response against *L. garvieae*. The progression of Lactococcosis in giant freshwater prawn occurs after the successful evasion of phagocytosis by *L. garvieae*. Up until now there is no prevention strategy described to *L. garvieae* in giant fresh water prawn. Also, for effective treatment there is a lack of knowledge of virulence factors and antimicrobial resistance of *L. garvieae* in giant fresh water prawn. Therefore, it is extremely important to investigate on the pathogenicity and innate immune response elicited during infection from *Lactococcus garvieae* in giant fresh water prawn.

Keywords: Aquaculture, *Macrobrachium rosenbergii*, *Lactococcus garviae*, Haemocytes, Immune response, pathogenesis

Short Biography

Dr. Omkar is an assistant professor at National Pingtung University of Science and Technology, Pingtung, Taiwan. He received his PhD in Aquatic Animal Health from National Pingtung University of Science and Technology. Dr Omkar completed his Masters in Aquaculture from Karnataka Veterinary Animal and Fisheries Science University, College of Fisheries, Mangalore, India. He then worked at University of Udine, Italy and University of Ghent, Belgium as a postdoctoral research fellow. Dr. Omkar has authored several publications in various journals and books. His publications reflect his research interests in fish and shell fish immunology, Aquaculture health management.

Noni, *Morinda citrifolia* fruit extract as potential antibacterial and increasing resistance of *Penaeus vannamei* postlarvae against *Vibrio parahaemolyticus*

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Introduction: Whiteleg shrimp, *Penaeus vannamei* is one of the most important shrimp species in the global aquaculture food production sector due high demand. Its fast growing nature and high tolerant to extreme water conditions makes this species a popular species to be cultured and produced. Acute Hepatopancreatic Necrosis Disease (AHPND) causing *Vibrio parahaemolyticus* was reported to largely affecting shrimp production due to mass mortality. The research on plant extracts as potential alternatives to conventional drugs against shrimp diseases such as antibiotics and chemical has been increasing for they are considered to be safer for both consumer and environment. Noni is recognised as one of the most important traditional medicinal Polynesian plant which had been used traditionally as therapeutic agent against illnesses among human such as diabetes, high blood pressure, muscle aches, poor digestions and many more.

Methodology: The extracts of *M. citrifolia* fruit was obtained through a series of process from drying, methanol extraction and rotary evaporation. There concentrated extracts were used for 3 separate experiments. Firstly, they were subjected to Gas Chromatography Mass Spectrometry (GCMS) for its bioactive compounds analysis. The second part of the experiment involved in the investigation of its antibacterial potential against *Vibrio parahaemolyticus* through *in vitro* antibacterial test in comparison with two other antibiotics. The third part of the experiment was the supplementation of the extracts into shrimp at different concentrations to determine its potential in increasing the survival of *P. vannamei* PL during challenge test with *V. parahaemolyticus*. The analysis on the hepatopancreas after the challenge test was conducted through histopathology.

Results: GCMS results showed a total of 45 bioactive compounds present in the fruit extract of *M. citrifolia* with Cyclononasiloxane, octadecamethyl- as the most abundant covering 36% of the total area in the GCMS peak chart. The *in vitro* antibacterial test showed that the extract was as effective as tetracycline against *V. parahaemolyticus*. The extract also showed its potential as an alternative to ampicillin against *V. parahaemolyticus* for ampicillin failed to inhibit the growth of *V. parahaemolyticus* as both tetracycline and the extract. The supplementation of the extract successfully increased the survival of *P. vannamei* PL up to 26.7% during the challenge test with *V. parahaemolyticus*. From the histopathology study, the presence of hepatopancreatic cells (B, R and E cells) were found to be higher in live shrimp of higher supplementation groups. The hepatopancreatic cells were completely absent in dead shrimps. The tissue structure of infected shrimps were analysed with distinct disruptions of tubules, connective tissues and haemocytes infiltrations among live shrimps of different supplementation groups as well as in dead shrimps.

Conclusion: The results from this study thus proved the ability of *M. citrifolia* fruit extracts as potential alternative for both antibiotics and diet supplementation as future aquaculture approach in increasing shrimp production and quality.

Sustainable extraction of natural immunostimulants from shrimp waste

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The growth rate registered by the seafood processing industries over the past years has been considerable. This could be explained by the health benefits associated with seafood products and the consequent increasing demands. However, this sector similar to other food processing industries generates a large quantity of waste. Hence, it is imperative to design and implement innovative solutions to mitigate the associated environmental burdens and maximize economic revenues. Crustaceans' wastes (e.g., shrimp shells) represent biomass possessing high content of minerals (20-50 wt.%), chitin (15-40 wt.%), proteins (20-40 wt.%), and some amounts of lipids and pigments, which are potential candidates to generate a wide range of high value-added chemicals. Chitin and chitosan are widely investigated as dietary additives with established beneficial biological properties, especially as immunostimulants. This study is focused on the sustainable extraction of chitin and chitosan from shrimp waste. This method of shrimp waste valorization helps to improve the productivity of fisheries as a result of the improvement of the aquaculture feed quality.

Keywords: Extraction; Natural immunostimulants; Chitin; Chitosan; Shrimp waste

Sustainable shrimp management: The critical role of advanced sustainability assessment tools

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Shrimp production is on the rise globally and the industry is being developed increasingly. Past events highlight the need for taking proper measures to ensure sustainable management of the shrimp farms is implemented effectively. This would bring about various advantages including more efficient use of resources, higher economic viability and resilience of the farms. These would in turn play an important role in more effective control of diseases. In order to analyze the sustainability features of a given farm, various advanced sustainability assessment tools can be applied. Those include life cycle assessment (LCA), exergy, and their combination, namely, exergoenvironmental analysis. The results obtained using these techniques can facilitate decision-making and future plans.

***Vibrio* species identified as possible component causes of shrimp white feces syndrome associated with the microsporidian *Enterocytozoon hepatopenaei* (EHP-WFS)**

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EHP-WFS was characterized by shrimp with white midguts that produce white fecal strings containing sloughed hepatopancreatic cells, tissue debris, bacterial cells and massive quantities of spores from the microsporidian *Enterocytozoon hepatopenaei* (EHP). Shrimp with EHP-WFS also exhibit loose exoskeletons, reduced feeding and retarded growth, high size variation, elevated feed conversion ratios and sometimes mortality. To investigate the possibility of the cause of white feces syndrome (WFS) is a pathobiome that includes a eukaryote (EHP) and bacteria (unknown species), using a high-throughput 16S rRNA amplicon sequencing analysis of bacterial microbiomes to compare the HP and midguts of WG and NG shrimp. The results revealed that principal-component analysis (PCA) and non-metric multi-dimensional scaling (NMDS) of WG and NG samples had different bacterial profiles and a significantly lower alpha-diversity was observed in WG than in NG samples by Chao1' s Richness index and Shannon's diversity index. For the bacterial taxa associated with EHP-WFS shrimp, Amplicon sequence variants (ASVs) of the genera *Vibrio* and *Propionigenium* were found with significantly higher-fold changes across WG samples than NG samples. The results from the laboratory immersion challenge test followed the method described by Aranguren et al., (2022) confirmed that the combination between *Vibrio* spp. and EHP to cause shrimp WFS under laboratory condition. EHP infected shrimp were continuously immersed with *Vibrio* strains; *V. parahaemolyticus* (non-AHPND strain), *V. parahemolyticus* (AHPND strain) and *V. campbelli*. White gut content has been initially demonstrated at 3 dpi for all EHP+*Vibrio* challenged groups, but not in the control group (EHP+broth). The overall results indicated that the combination of EHP and *Vibrio* infection could induce WFS in shrimp. We hypothesize that shrimp loss appetite during their course of EHP infection, resulted in the leftover of feed for bacterial overgrowth. Consequently, the combination of EHP with *Vibrio* infection induced WFS in shrimp.

Keywords: White feces syndrome, *Vibrio* spp., *Enterocytozoon hepatopenaei*, *Penaeus vannamei*

Short Biography

Dr. Natthinee Munkongwongsiri is currently working as a Postdoctoral researcher at Aquatic Animal Health Research Team, BIOTEC, NSTDA, Thailand. She received her Doctoral degree in 2016 from Kasetsart University, Thailand. Her research interest is in topics of shrimp diseases and disease management. Her expertise is in shrimp histology and pathology.

Effect of oral administration of *Zingiber officinale* extract on growth and immune functions of shrimp *Penaeus monodon* against white spot syndrome virus (WSSV)

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WSSV has emerged worldwide as one of the most prevalent and widespread viruses in shrimp aquaculture. The virus currently causes the most damage to the global penaeid shrimp production, leading to massive mortality (80 to 100%) within 3-10 days. *Zingiber officinale* is one of the most commonly and extensively used plants in various traditional and folk medicines throughout the world. We examined the effect of an extract of *Z. officinale*, orally administered with feed, on growth and immune functions of black tiger shrimp *Penaeus monodon* against WSSV. The plant extract was incorporated in artificial pellet feed at a concentration of 0% (control), 0.05 % (Treatment 1, T1) and 0.1% (Treatment 2, T2), and fed to the shrimps for 4 weeks. After the feeding trial, each group of shrimp was challenged with WSSV by injection, and was monitored for two weeks. WSSV-infection was confirmed by PCR after challenge of shrimp, and qPCR was used to examine gene expression. Growth performance (final body weight, weight gain, specific growth rate (SGR)) and feed utilization ((low feed conversion ratio (FCR), high protein efficiency ratio (PER)) of shrimp fed with extract T1 were significantly ($P < 0.05$) improved compared to shrimp fed with the basal diet. All the immunological parameters i.e. total haemocyte count (THC), prophenoloxidase activity (proPO), and superoxide dismutase activity (SOD), were increased significantly ($P < 0.05$) in extract-treated shrimp compared to control, and the haemolymph clotting time (HCT) was decreased significantly in treated shrimps both in pre- and post-challenge conditions. Before challenge, no significant difference was found in the survival of shrimp, but after challenge, the cumulative mortality was significantly lower in T1 (20%) and T2 (75%) than in the controls (100%) 14 days after challenge. In conclusion: oral administration of *Z. officinale* extract can enhance the growth and immunity as well as resistance capacity against WSSV infection, and PCR can be used as a diagnostic for experimental WSSV infection.

Keywords: *Zingiber officinale*, Growth, Immunity, WSSV, *Penaeus monodon*, PCR

Short Biography

Mr. Alokesh Kumar Ghosh is doing his PhD under the department of Biology at KU Leuven and also acting as an associate professor at Fisheries and Marine Resource Technology Discipline of Khulna University, Bangladesh. He has completed his MSc on Aquaculture and Marine Resource Management from the Wageningen University of Netherlands. He has a broad experience in shrimp farming, shrimp immunity, their disease producing pathogen and inhibiting the pathogen using plants and probiotics. He has more than 20 numbers of peer reviewed publications.



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Usaha Fadzilat (M) Sdn Bhd was incorporated in 1987, by Mr. Lim Tio Huat, a visionary and a persevering industrialist who had a hope to produce more quality and healthy food from marine resources, for more people to enjoy. Mr. Lim started with 13 ponds over 70 acres of land to cultivate tiger prawns. From there he further ventured into the rearing of pacific white shrimp, as he gradually expanded his business as a result of his quality products and fine response from the market. The company, which has already brought in the new fish fry from Taiwan in the year 2014, was very focused in the R&D and cultivation of MilkFish (also scientifically known as Chanos chanos), for the two subsequent years.



EMPOWERSTYLE

Zumail Global Sdn. Bhd.
Suite 20.07, Wisma Zelan, Jalan Tasik Permaisuri 2
Bandar Tun Razak, 50000 Kuala Lumpur

Tel: Main: +60-14 253 9897
Email: zumail.global@gmail.com
Web: <https://www.zumailglobal.com>
Contact Person: Mrs. Mazura Binti Abdul Aziz

ZGSB is a combination of a group of individuals with over 22 years of experience in various fields of background especially PUBLISHING, PRINTING & supply of CORPORATE GIFTS including branded merchandise. ZGSB also has strategic business partners in the field of MEDIA TECHNOLOGY such as Artificial Intelligence (AI) & Augmented Reality (AR). We also get direct supply support from Malaysia's leading factories for corporate gift and branded merchandise. ZGSB will FOCUS on helping to provide friendly services such as government agencies, GLCs and SME Entrepreneurs especially to get commercial value quality and trendy prints & corporate gifts, within the stipulated period.



Blue Archipelago Berhad
No. 3, Jalan Perindustrian Puchong
Bandar Metro Puchong, 47160 Puchong Selangor

Tel: Main: [+603-8074 7555](tel:+603-80747555) | [+60 12-273 3483](tel:+6012-2733483)
Email: marketing@bluearchipelago.com
Web: <http://www.bluearchipelago.com>
Contact Person: Mr. Wu Wai Leong

Pelagos is a brand of products from Blue Archipelago which can be assured of its highest quality and uncompromising dedication to produce safe and sustainable seafood. The winning edge of our Pelagos product: 4-star product traceability, 100% Hormone and Antibiotic FREE, Seawater farmed for tastier shrimps, quick frozen for freshness, accorded with HACCP, BAP and EU Certifications that meet stringent global standards. Email us at marketing@bluearchipelago.com for more information.



QS Tech Sdn. Bhd.
InnoHub Putra Science Park UPM
UPM Jalan Maklumat,
43400 Serdang, Selangor

Tel: Main:+60 14-3376414
Email: qstech@gmail.com
Contact Person: Ms. Maya Liyana Hamzah

QS Tech Sdn Bhd is funded by the International Development Research Center (IDRC, Canada) together with Department of Health and Social (DHSC, UK) and Innohub UPM in 2021. This company aims to develop additives for disease control through quorum quenching technology. This technology inhibits bacterial virulence through specific signal quenching that hinder the interactions between the pathogenic bacteria. The use of the QUENCH product as an alternative to antibiotics can minimize the dependency on antibiotics in shrimp aquaculture and slow the emergence of multidrug-resistant (MDR) bacteria.



S.R.I H.I.J.R.A.H (M) S.D.N. B.H.D

Sri Hijrah (M) Sdn. Bhd.
70-3-51, D Plaza Mall
Bayan Baru, Penang, 11900 Pulau Pinang

Tel: Main:+60 10-401 0000
Email: srihijrah@yahoo.com
Contact Person: Mr. Izrol

We are a wholly owned bumiputra private company with main core business specialized in supplying Scientific Equipment, Scientific Apparatus, Laboratory Equipment, Laboratory Furniture, Medical Disposables for School of Science, Education Equipment, Marine Equipment and Office Stationery and Equipment.



Caya Primer Sains Sdn. Bhd.
Suite 3.1, Level 3, UmXcelerate
University of Malaya, 50603, Kuala Lumpur

Tel: Main: +60 10-401 0000
Email: intanshazlin57@gmail.com | info@cayaprimersains.com
Contact Person: Ms. Intan Shazlin Binti Tajul Ashikin

CAYA PRIMER SAINS SDN BHD was incorporated on 1 April 2011. The scope of our company is research, development, services and manufacture of molecular diagnostic detection kits. We are dedicated to providing quality and accurate diagnostic, proper and particular services, technical and management services to our customers. We will strive to implement a long term relationship with our clients, based on safety, quality, timely service and an expectation of their needs.



F1 Aquaculture Sdn Bhd
No. 15, Lorong Enggang
Taman Keramat 54200, Kuala Lumpur

Tel: Main: +60 13-602 5577
Email: f1aqua@itcare.com.my
Contact Person: Mr. Tan Jia Hoe

Our company is a hatchery that produces marine fish seeds such as grouper, red fish, sea bream and others. A modern indoor seeding system is used to produce healthy and quality fish seeds. With the practice of the traceability system and the myGAP scheme, F1 Aquaculture Sdn. Bhd. thrives to produce fish that is safe to eat.



Malaysian Frozen Foods Processors Association
4572, Jalan Chain Ferry
12100 Butterworth, Pulau Pinang

Tel: Main:+60 12-4221178
Email: mffpa@ymail.com
Contact Person: Jessie Lim

TANGAWIRA SDN. BHD.

Tangawira Sdn Bhd
Jalan Sungai Karang, 5km Off Jalan Sikuati
89058 Kudat, Sabah, Malaysia

Tel: Main:+60 16-588 6616
Email: tangawira@outlook.com
Contact Person: Lee Chin Fah

TANGAWIRA SDN. Ltd. is a factory that processes fish fertilizer or FISHMEAL that was established in 1998 in Kudat district, Sabah. The factory is located at Jalan Sungai Karang, 5km off Jalan Sikuati, 89058 Kudat, Sabah. The factory has more than 75 employees and is equipped with effective facilities. The factory operates 24 hours a day and has two shifts. The establishment of this company is to produce fishmeal as supplementary food for farm animals such as chicken, fish, cattle and so on. In addition, this additional feed can also provide guaranteed health to farm animals and farmers can produce quality livestock feed.



Hong Cheng Seafood Supplies Sdn. Bhd.
Lot 7297, Jalan Perusahaan 2
Kawasan Perindustrian Parit Buntar
34200, Parit Buntar, Perak

Tel: Main: +60 5-716 6899 | +60 12-578 4899
Email: hongchengseafood@hotmail.com
Web: <https://www.hongcheng-seafood.com>
Contact Person: Khor Chiew Yong

Hong Cheng is an excellent seafood supplier. It first opened its doors in December 2008 and is run and co-owned by Chiew Peng and Chiew Yong. In 2008, "Hong Cheng seafood supplies sdn. bhd" was officially founded in a fish village in Penang. We started with an operation space not bigger than half a basketball court. However, we are determined to grow and become the best seafood supplier company, selling the best quality seafood products.



MSAVE MARKETING SDN BHD

Msave Marketing Sdn. Bhd.
17-G, Jalan LP 7/4F
Taman Lestari Perdana
43300, Seri Kembangan, Selangor

Tel: Main: +60 3-80803635 | +60 11-65217400
Email: msavemarketing.com.my
Contact Person: Umi Haizan Binti Abd Karim

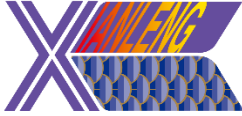
Package - Shrimp Sponsors Company Profiles



Dragon Taste Seafood Export & Import Sdn. Bhd.
No.73, Jalan Selat Selatan 7/KS05,
Taman Perindustrian Sobena Jaya,
42000 Pandamaran,
Port Klang, Selangor, Malaysia

Tel: Main:+60 12-705 3691
Email: comm@xianleng.com.my
Web site: <http://www.dtsgroup.com/>
Contact Person: Lee Yee Jing

SeaNutri is an innovative brand created under Dragon Taste Seafood Wholesale & Retail Sdn Bhd (DTSWR). DTSWR is the first company established by DTS Group, and served as an important foundation for the group. With the creation of SeaNutri, it marks an important development milestone for DTS. At the starting point, wholesale distribution plays the main distribution activities for SeaNutri. It is distributed widely to hotels, restaurants, hospitals, high-end eateries, etc. Retail distribution started actively in recent years. It penetrates into the market by reaching out to the end consumer with its various choices of products from fresh and frozen fish, Ready to Cook, Ready to Eat, Ready to Heat, etc. Over the years, SeaNutri has become a recognised premium fish brand in the hospitality industry. It is the most trusted brand among 5-star hotel chefs



Xian Leng Trading Sdn. Bhd.
No. 35, Jalan Penaja 3
8300 Batu Pahat, Johor

Tel: Main:+60 12-705 3691
Email: comm@xianleng.com.my
Web site: xianleng.com.my
Contact Person: Kuan Kai Seng

Xian Leng Trading Sdn. Bhd. was established in 1989 which is engaged in commercial captive breeding of the Asian Arowana, Stingray, Goldfishes, Cichlids, Anabantids fishes. The ornamental fish trading center is located at Batu Pahat, Johor, engages in the retailing of both local and exotic aquarium fishes as well as the related paraphernalia such as aquarium and accessories, fish feed and medication.



Gertak Sanggol Hatchery Sdn Bhd
94-A, Mukim 9, Jalan Gertak Sanggul
11920 Teluk Kumbar, Penang

Tel: Main: +60 13-686 3535
Email: comm@xianleng.com.my
Web site: xianleng.com.my
Contact Person: Kuan Kai Seng

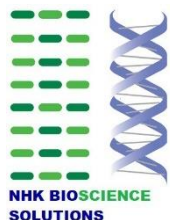
Gertak Sanggol Hatchery Sdn Bhd (GSHSB) was established and has been operating since 18 years ago in 2004 until now and the hatchery is located at Pulau Pinang, Pahang, and Sabah. Our company's main activities are supplying nauplii and post larvae for both P. Vannamei shrimp and P. Monodon shrimp throughout Peninsular Malaysia including Sabah and Sarawak. GSHSB was recognized and awarded MyGap and FQC certificates by the Department of Fishery Malaysia (DOF) since 2013 until now.



QL Fishmeal Sdn. Bhd.
Lot 164, 2647 & 3314,
Jalan Tepi Sungai
36400, Hutan Melintang, Perak

Tel: Main: +60 16-230 9648
Email: Inquiry@qlfishmeal.com
Web site: <https://www.qlfishmeal.com/>
Contact Person: Ms. Lim Siew Lian

QL Fishmeal Sdn. Bhd. has been established since 1991 to provide high quality fishmeal by utilizing "Steam-Drying" technology for both the local & international markets. We are located near a fishing village, named Hutan Melintang, which enables us to obtain fresh raw fish directly from the fishing vessel every day. Our fishmeal is freshly made from 100% whole fish and bones/offal from processed fish, with a trace amount of natural sea creatures. High freshness of fishmeal is maintained by completely processed the in-coming raw material within 24 hours of upon receiving.



NHK Bioscience solution Sdn. Bhd.
27-A, Lebuhraya Gelugor
Georgetown 11600 Penang

Tel: Main: +60 17-336 2192 | 03-7987 8218
Email: geehean@nhkbioscience.com
Web site: <https://www.nhkbioscience.com>
Contact Person: Kuan Kai Seng

NHK Bioscience Solutions Sdn. Bhd., located in Kuala Lumpur, Malaysia, was established in June 2003 and commenced operations in August 2003 as a Life Science company supplying products and services to the scientific, (bio)medical and biotechnology communities engaged in molecular research and/or diagnostics. To the communities whom we serve, we pay careful attention to our customers' needs and endeavour to meet them by offering the latest cost-effective technological solutions. We aim to achieve a high degree of customer satisfaction and develop trust through offering quality solutions matched by excellent technical support and exceptional service.



Hai Leng Enterprise Sdn. Bhd.

Sedco Light Industrial Estate, Phase 1,
Lot 1, 2, 10 & 11, Mile 3, North Road,
90000 Sandakan, Sabah

Tel: Main: +60 89-211 713 | +60 89-217 779

Email: hailengsdk@gmail.com

Contact Person: Kuan Kai Seng

"Hai Leng Enterprise Sdn Bhd has been a HACCP certified seafood company since 1984. It is one of the leading seafood processing companies in Sandakan, Sabah, Malaysia. Due to the advantage of its natural environment, the ocean surrounding Sandakan, Sabah is one of the places in Malaysia producing the best quality of seafood. We also produce and supply our own farm-raised aquaculture shrimps with great quality in freshness, taste, and color appearance."



Syaqua Sd. Bhd.

No.19 Jalan Perigi Nenas 7/2 Ks 11,
Kwsn Perindustrian Pulau Indah

Tel: Main:+60 14-955 0146

Email: gabreal.j@syaqua.com

Web site: www.syaqua.com

Contact Person: Gabreal Johnny Gabil

SyAqua is the global leader in genetics and early nutrition for aquaculture. We aim to create optimal balance genetics focusing on speed of growth and good survival rate. With SyAqua's Balanced Line, we use the latest technologies in genomic selection, quantitative genetics, and breeding science to significantly improve the genetic response to ever-challenging aquaculture climates and environments. This can enlarge the population of successful shrimp farmers benefiting from SyAqua's total solutions with greater yield and faster growth.



*"Innovative Technological Solutions for
Our Spatial World"*

Spatialworks Sdn. Bhd.
Blok C-T.01-U.04, No.1 Jalan P8D
62250, Putrajaya

Tel: Main:+60 3-8881 0962
Email: admin@spatialworks.com.my
Web site: <https://www.spatialworks.com.my>
Contact Person: Cik Nurnajmi Binti Ibrahim

Spatialworks Sdn. Bhd. is a technology driven GIS-IT and environmental consultancy company, provides a complete spectrum of geographic information system (GIS) and technology driven environmental solutions to both public and private sector clients. Spatialworks Sdn Bhd is a client-focused company where it is our commitment to provide cost-effective solutions for the clients, continuously seeking for the best and the latest state-of-the-art technological solutions for the betterment of our environment, and to support them from beginning to the end in their efforts. Our staff is a group of highly focussed and dedicated professionals who have been working together with proven commitment to our clients in delivering above expectation services and products.



Uni-President

Uni-President (M) Sdn. Bhd.
3A-13, Blok A, Damansara Intan,
No 1, Jalan SS 20/27
47400, Damansara, Selangor

Tel: Main:+60 16-916 1001
Email: uni.president.malaysia@gmail.com
Web site: <https://uni-president.com.vn>
Contact Person: Mr. Benny Leng

Uni-President Enterprises Corp (UPEC), one of the biggest food groups in Taiwan and the top 10 players in China & Asia. UPEC has over 100 subsidiaries and affiliates worldwide. We are committed to providing the finest products and quality services that are best suited for customers. Good quality, Good credibility, Good services and Fair price are the quintessential management philosophy of Uni-President, and it has become the cornerstone of Uni-President Enterprises Corp. Uni-President Enterprises Corporation embodies the management philosophy of integrity, Diligence, Innovation, and Progress to the Future.



Shrimp Improvement System PTE
16 New Industrial Road 05-03/04
Hudson Technocentre Singapore
(536204)

Tel: Main: +65 9781 9454
Email: Edmund_Koh@shrimpimprovement.com.sg
Web site: <http://www.shrimpimprovement.com/>
Contact Person: Koh Wei Lun Edmund

Founded in 1998, Shrimp Improvement Systems (SIS) is the world's leading provider of shrimp broodstock. SIS produces genetically improved and Specific Pathogen Free *P vannamei* shrimp broodstock. Our breeding facilities are currently located in Florida, Hawaii. We are also the major supplier of the best-performing broodstock to external shrimp farming operations throughout Asia and the Americas. As the premier shrimp breeding company, SIS is dedicated to improving the performance of aquaculture stocks worldwide.



Global Goodway Sdn. Bhd.
125 R & S, Desa Tanjung
Jalan Tanjung Tokong
10470, Georgetown, Penang

Tel: Main: +60 16-400 5316
Email: mike.saw84@gmail.com
Web site: <https://www.globalaquaculture.asia/>
Contact Person: Michael Saw Zng Hoz

Global Goodway specializes in Tiger Prawn Farming, with our high volume production methodology and continuous improvements, we are able to consistently produce Tiger Prawns of the highest Quality. Since 2019, we've also ventured into IoT with the objective of automating key processes and providing management with continuous data. Our latest R&D project, LuFloc, a super-intensive system with minimal water exchange has proven to be a success and it is already in the production stage.

Package - Seabass Sponsors Company Profiles



Biogenes Technologies Sdn. Bhd.
Block 2, Infrastructure University Kuala Lumpur,
43000 Kajang, Selangor

Tel: Main:+60 12 6656 360
Email: contact@biogenestech.com
Web site: www.biogenestech.com
Contact Person: Tang Kok Mun

Biogenes develops and manufactures digital-based diagnostics products for diseases in human and animal healthcare, and testing solutions in food safety as well as pollution monitoring. Our app-based solutions allows doctors and farmers alike to upload their test results together with time and location data into a cloud-based server for real-time and remote access from anywhere in this world. Biogenes collaborates closely with research institutions and universities to jointly develop these solutions as well as conduct field trials to validate our technologies. Our collaborators include, among others - MARDI, DVS and FRI.



Freecap Resource Sdn. Bhd.
Lot T-5, Lumut Port Industrial Park,
Kampung Aceh Mukim Lumut,
32000 Sitiawan, Perak, Malaysia

Tel: Main:+60 12-616 8911 | +60 5-691 0850
Email: bw.ng@platinum2u.com
Web site:<https://pcfgroup.com.my>
Contact Person: Dr. Ng Bee Wah

Freecap Resource Sdn. Bhd. (FRSB) was founded in association with PCF group, a group with more than 30 years old involvement in Malaysia sea catch seafood industry. FRSB is a one-stop seafood supply chain premise certified with HACCP, GMP, MeSTI and HALAL. We are specializing in crustacean products (*Penaeus monodon*) to fulfill the global market needs as well as the local market. With the sufficient capital, professional experience and strong scientific support team giving FRSB a renewed future in producing sustainable seafood to secure food security.



Helping the world *thrive*

Cargill Feed Sdn. Bhd.
Lot 55711, Dry Bulk Terminal, Jalan Mawar
West Port, 42009 Klang, Selangor

Tel: Main: +60 16-410 3234
Email: Sankari_Ranjapan@cargill.com
Web site: <https://www.cargill.com.my>
Contact Person: Mrs. Sankari A/P Ranjapan

Cargill has been conducting business in Malaysia since 1978. Today, our businesses include vegetable oil refining and production of value-added products, grain and oilseeds distribution, animal nutrition, sales and marketing of starches, sweeteners and texturizers as well as cocoa and chocolate products. Headquartered in Kuala Lumpur, Cargill currently employs about 740 people in 7 locations countrywide.



PENAWAR INTERNATIONAL

Penawar International
No. 32-1, Jalan jade Hill Utama
Commerce Village
Jade Hills, 43000 Kajang, Selangor

Tel: Main: +60 3-8740 0799 | 13-368 9502
Email: salespenawar@gmail.com
Contact Person: Mr. Joyhanis Bin Mohd Jamil

PENAWAR INTERNATIONAL SDN BHD was established in January 2020 with the Malaysian Companies Commission bearing registration number 1356487-P. Penawar International Sdn Bhd is registered in Selangor, Malaysia and is 100% bumiputera owned. Penawar International Sdn Bhd is actively involved in Exhibitions, Events, Galleries, and Museums. This company has been heavily involved in business based on "Event Management". This includes organizing parties, event management, cultural performances, dinner parties, organizing sports and family days. Through this business activity, the company has established many business networks with audio services, catering, event tools such as canopies, interior decorations and others.



Fandi Ads Resources
No. 17-2f Jalan Setia Perdana BA U13/BA Setia Alam
Seksyen U13
40170 Shah Alam Selangor Darul Ehsan

Tel: Main: +60 12-066 0878
Email: fandiadsevent@gmail.com
Contact Person: Mr. Affandi bin Daud

We transform even the simplest of ideas into innovative and well-crafted products that fit the needs of our clients and exceed their expectations. We offer outstanding value to our clients by providing the highest quality services at very reasonable investment. Our aim is to build a loyal patronage by providing our clients with quality, reliable end products and as well as efficient, friendly customer service. @FANDI ADS.... we constantly strive to challenge the conventional methodology in current trends in order to deliver the best and effective solutions for a given project.



Canvio Sd. Bhd.
No. 22-1, Jalan Setia Utama AU, U13/AU,
Seksyen U13, Setia Alam,
40170 Shah Alam, Selangor

Tel: Main: +603 3341 4392 | +6014-3314780
Email: info@canvio.net
Web site: <https://www.canvio.net/>
Contact Person: Stephenie Poh (stephenie@canvio.net)

Canvio Sdn Bhd is a comprehensive solution provider for the research and diagnostic market in Malaysia. Canvio is led by a group of dynamic professionals and over the years, we have assisted countless clients in achieving their research and diagnostic outcomes while encouraging the adoption of molecular biology and cell therapy in a forward direction. We consciously increase technology awareness by organizing numerous seminars, trainings and workshops on molecular science and its applications to various customer segments; in academia, in clinical diagnostics and also to the industry.

Package - Oyster Sponsors Company Profiles



Kaizai Resources
No. 7, Bangunan MDY, Jalan Pantai Murni
Gurun 06900, Kedah

Tel: Main: +60 12-411 8337
Email: kaizai.kay@gmail.com
Contact Person: Mr. Md. Khir Johari Bin Md. Yusoff

KAIZAI RESOURCES ada sebuah syarikat yang melibatkan diri didalam industri Peralatan Saintifik, Penyelidikan, Pharmaceutical dan Hospital. Di tubuhkan pada 30hb. Disember. 2008. KAIZAI RESOURCES telah menyahut saranan yang dibuat oleh Kerajaan Malaysia untuk bersama –sama memajukan negara didalam bidang tersebut. KAIZAI RESOURCES adalah syarikat yang dimiliki oleh 100% Bumiputera.



UD Technology Sdn. Bhd.
Plot 80, Tingkat Perusahaan 6
Kawasan Perusahaan Prai 4
13600, Penang Malaysia

Tel: Main: +60 4-501 2231 | +60 13-786 4191
Email: udtechnology@gmail.com
Web: ud-technology@gmail.com
Contact Person: Mr. Patrick Teoh

Founded in 2008, UD Technology Sdn Bhd specializes in laboratory equipment and analytical supplies. With our high quality products, competitive pricing, excellent technical and application support contributed to UD Technology Sdn Bhd, quick growth and the territory was expanded through the global network. We can offer a wide range of products to meet your chromatography needs from sample preparation to analysis in a variety of fields e.g. pharmaceuticals, foods, drugs, chemicals, petrochemicals, environmental, natural products, biotechnology, etc. We are also very proud of our full technical or application support and guarantee for customer satisfaction. We are willing to customize our products to meet any specific requirement of our customers.

KAULUAN SDN. BHD.

Kauluan Sdn. Bhd.
F-2-15, Blok F, Tingkat 2,
Lorong Inanam Kapital 5, Inanam Kapital,
88450 Kota Kinabalu, Sabah

Tel Main: +60 17-823 3218
Email: kauluansb@yahoo.com
Contact Person: Mr. Alex siaw

Kauluan Sdn Bhd is a certified fishmeal manufacturer based in Kota Kinabalu, Sabah. We produce high quality fishmeal with crude protein more than 60% for the international market. We are dedicated to maintaining high quality and freshness of our products by processing the raw fish from fishing boats immediately upon receipt daily. We also send our fishmeal samples for our in-house lab and external lab for tests to ensure standards and requirements are met.



Jeit Yit Hardware & Household
No. 15, Jalan Deluxe, Pusat Perdagangan Deluxe
Dengkil, 43800 Selangor

Tel Main: +60 12-360 0052
Email: jeityeit@yahoo.com
Web: <https://jeityeithardwarehousehold.business.site>
Contact Person: Lee Chin Seng

We are selling a variety of quality hardware tools including Seamaster and Kansai paints, Black Hammer safety shoes, duplicate keys and car immobilizer, a variety of cutting meat knives and so on... Do contact us for any queries.



Australia

Yuko Hood

Bangladesh

Mohammad Shamsur Rahman
Shafiq Rheman

Belgium

Alokesh Kumar Ghosh

Brunei

Abdul Salam Wafi Haji Md. Diah
Muhammad Abdul Hakeem Julaihi

China

Qiyao Wang

Chinese Taipei

Omkar Vijay Byadgi
Ramya Kumar
Cong-Yan Chen
Yen Siong Ng

Yu Kent Tan
Grace Chu-Fang Lo
Han-Ching Wang

France

Jerome Delamare-Deboutteville

Greece

Mr. Gerald N Misol Jr

India

Neeraj Sood
Syed Syariq Nazir Qadiri
Pravata Pradhan

Indonesia

Agus Sunarto
Rajeev Kumar Jha
Nugroho Wiratama

Dewi Syahidah
Winarti Achmad Sarmin Djainal

Iran

Reza Heydari

Italy

Melba G. Bondad-Reantaso

Japan

Motohiko Sano
Akito Hayashi
Hiroaki Saito

Malaysia

Abdul Hafis Abdul Rashid
Amirah Fatimah Md Nordin
Aznaliza Yahya
Ching San Tan

Azmar Hana Elliany Azhar
Mushidi Hassan
Mustafa Asmuni
Nata Bakar

Azahari Othman
Adnan Hussain
Chew Poh Chiang
Ahmad Daud Om
Ahmad Syafiq Ahmad Nasir
Andrea Lim Li Li
Azhar Hamzah
Azila Abdullah
Chadag Vishnumurthy Mohan
Chen Fei Low
Chiun Khang Yong
Erniyta Osman
Hajar Rastegari
Haslawati Baharuddin
Hidayah Manan
Hirzahida Mohd Padil (record not found)
Hock Tieng Chuo
Hon Jung Liew
Hung-Hui Chung
Ina- Salwany Md Yasir
Jackie Peter
Jeffry Ling
Jerome Delamare-Deboutteville
Julia Hwei Zhong Moh
Kua Beng-Chu
Maisarah Ab Samad
Mohamad Nor Azra Md Adib
Mohammad Munir
Mohammad Noor Amal Azmai
Murni Karim
Natrah Fatin Mohd Ikhsan
Naziah Muntil
Ng Wing-Keong
Noor Affizah
Nor Azman Kasan
Norhafiza Ilyana Yatim
Nur Athirah Mohd Jazman
Nur Fatimah Abd Halid
Nur Nazifah Mansor
Padilah Bakar
Reza Heydari
Rimatulhana Ramly
Roslianah Asdari
Roslina Ahmad Nawawi
Samsur Mohamad
Sandra Catherine Zainathan
Shaharah Mohd Idris
Shuhadah Mustapha
Siti Norita Mohamad
Sylmie Al-Harir
Tak Seng Leong
Tilusha Manchnayake
Venmathi Maran Balu Alagar
Putri Nurul Sikira Mustaffa
Aida Nabilla
Waiho Khor
Wan Norhana Md Noordin
Mohd Firdaus Naw
Abdul Hafiz Mohd. Zaki
Abdul Hakim
Abdul Halim Yousof
Abdul Rahman Ahmad Pauzi

Nazri Ishak
Ng Kooi tiam
Pang Anak Nyukang
Nik Ahmad Tarmizi Nik Ismail
Nik Nazli Effendy Ramli
Nordiskandar Nor Tajudin
Nummeran Mohd Nordin
Nurassyaima Kamari
Nurdin Shafie
Osman Hamzah
Perceval Conder
Raja Yana Meleessa Raja haron Arashid
Rizman Zulkarnaen Suardi
Rizwan Nordin
Roslan Abu Hasan
Roslee Dela Cerna
Rosli Ismail
Rudy Zaidi
Ruzaidi Mamat
Ryanto Saifuddin
Saadon Kasmon
Sabda Safiee
Saiful Azuan Jamalludin
Shafiq Rheman
Shahidan Hashim
Shahrul Naim Abd. Razak
Shahrul Nizam Baharuddin
Shallehuddin Abdul Taib
Shamsol Muhammad
Simen Mudin
Snehashish Snehashish
Sow Cyn Shieng
Sufian Ahmad
Sukri Deris
Syafiq Johar
Syafiq Syauqi
Syed Amirrudin Syed Suhaini
Syed Mohamad Azim Syed Mahiyuddin
Syed Yusuf Wan Drahman
Teoh Pik Neng
Tn. Mohd. Hakimi Tn. Mood
Wan Amizan Wan Ahmad
Yau Chang Siew
Zahaludin Amir Danial
Zaidil Abdilla Ahmad Salehuddin
Zainal Hanapi
Zakaria Che Din
Noraini Md Nor
Adillah Ibrahim
Afzan Muntaziana Mohd Pazai
Ainul Riza Abu Seman
Aminah bt. Ignasius
Amy Aineda Omar
Amy Noor Zaina Amir Hamzah
Anis Mazidah Abd Samad
Aqilah Mohd. Yusof
Aranja Fong
Azimah Jumatli
Azlina Apandi
Azura bt. Markus
Azwa Abdul Hamid
Chai Pui Shan

Abdul Razak Man
Abdul Razak Hamzah
Abdullah bin Jaafar

Abu Bakar Al-Siddiq Abd Salam
Abu Bakar Tumin
Abu Yazid Yusnizab Muhammad
Affan Nasruddin Roshidi
Ahmad Fauzi Mohd Khosim
Ahmad Helmi Hj. Chik
Ahmad Najmi Nasruddin
Ahmad Redzuan Ramli
Ahmad Rohaimae Abdullah
Ahmad Sabri Katiman
Ahmad Saifullah Mohammad
Aiman Zulkilee
Albert Apollo Chan
Alistair Patrick
Alvin Aldreen
Alvin Clinton Anak Suder
Amadi Tubah
Amar Fakar Adzuan Roslan
Amer Shahjehan Hassan
Amir Hamzah Ramli
Arifin Drahan
Arthur Besther Sujang
Aslah Mohamad
Asrani Abdullah
Azaharie Anuar
Azizul Abu Hasan
Azmi Rani
Azri Md Yusoff
Azrim Sulong
Bakri Saad
Basri Abd. Rashid
Basri Man
Benedict Iber
Brendan Tan Heng Wee
Buniamin bin Kiprawi
Che Shahidi Che Mod
Che Zulkifli Che Ismail
Chin Han Kiat
Chrysostom anak Thomas
Danial Iman Haris Nor Azma
David Yambun
Dokanaer Kasto Anak Muning
Dzulfikkar Baitul Ma'mur
Fairol Tajuddin Suhaili
Faizal Ibrahim Suhaili
Fakhruddin Bokhari
George Bobby
Hafiz Syahri
Mohd Sofi Yaacob
Hanan Mohd Yusof
Hasbullah bin Harun
Hasyaruddin Abdul Halim
Haziq Aiman
Hilal Abd Aziz
Darhan Nawawi
Zainudin Abd. Wahab
Iftikhar Ahmad Abdul Rafi
Irfan Hakimi

Cindy Kho Jia Yung
Dayang Zafirah Abang Jashmady
Doreen Wee Siew Leen
Emylin Badut
Erin Tan
Fadzilah Yusof
Faizah Nor Mohamed
Faridah Zulkefly
Haliza Sulaiman
Hanisah Azmi
Haryati Abd. Wahab
Hasaniah Ahmad Kamalluddin
Hasnisa Abdul Hamid
Hazreen Nita Mohd Khalid
Hemalatha Raja Sekaran
Hidaya Dmulliany Mohd Sidek
Iffah Nadhirah
Intan Nurlemsha Baharom
Jafrizah bt. Abd. Razak
Kamisa Ahmad
Keni Anak Ngijwol
Khaitrul Ediana Mohd Tahir
Khazlita Adzim
Kho Li Yung
Lim Ai Gaik
Liyana Ramli
Liza Long
Lourdemary George
Mariam Marip
Maznah Yusoff
Munirah Zainol Abidin
Muslihah Abdul Rahaman
Najihah Mohamad
Bakri bin Miswan
Noor Amira bt. Muslim
Noor Asma bt. Mohammad
Noor Hasmayana bt. Yahaya
Noor Ikhwanie Zainal
Noor Suhailis Zelani
Nor Asnisawati bt. Yusof
Nor Azlina Abdul Rahman
Nor Haida Ishak
Nor Khalilah bt. Zainuddin
Noor Mahya Yusof
Nor Mastura Hasan
Noraisah Zainuddin
Noraishah bt. Hashim
Noraisyah bt. Abu Bakar
Norarziah Abdullah@Aziz
Norashikin Anjur (record not found)
Norashikin Daliyana
Norasmah Mantali
Norhabina Abd. Aris
Noridayu Alias
Norma Said
Normadiha Mat Nor
Nur Amirah Nazirah Jasemi
Nur Aqilah Mohd Salim
Nur Ashikin Arbi
Nur Erna Liyana Adam
Nur Fadhilah Zainul Mustaffa
Nur Fatin Afifah Osman Manah

Irwan Paiman
J. Denwinner Jikon
Jackson Clive Jusak
Jamil Musel
Jidy Joseph
Johari Tim
Ka Hong Tee
Kaharudin Md. Salleh
Kamarul Anwar Mohamed Zabri
Kasim Tawe
Kok Mun Tang
Kon Yeu Hooi
Liew Vui Kien
Lindy Enggong
Lukman Basri
Luther Moguup John
Mamat Amin Abd. Rahman
Marzie Ryn Romin
Marzuki Morad
Mohamad Azlan Amran
Mohamad Fitri Arris
Mohamad Ikraf Mohamad Sharif
Mohamad Iskandar Ishak
Mohamad Nazri Puasa
Mohamad Sani Thalib
Mohamad Sufiyan Salmi
Mohammad Hafiz Hassan
Mohammed Roshaziat Mustaffa
Mohammed Suhaimee Abd. Manaf
Mohammad Kamruzzaman Hossain
Mohd Armia Mohd Salleh
Mohd Asmadi Alawi
Mohd Asraf Said Azman
Mohd Azhar Mohd Zin
Mohd Azrul Mahmud
Mohd Fazhan Mohd Hanafiah
Mohd Firdaus Ahmad Pauzi
Mohd Firdaus Azmi
Mohd Ghazali A Manap
Mohd Hariz Ab Halim
Mohd Hazrul Muhammad
Mohd Lazim Mohd Saif
Mohd Maliku Seman
Mohd Nizam Ismail
Mohd Nor Azman Ayub
Mohd Nor Isyam
Mohd Nur Aminullah Abu Bakar
Mohd Ridzuan Mohamad Mokhtar
Mohd Samsul Rohizad Maidin
Mohd Shafiq
Mohd Syafiq Mohammad Ridzuan
Mohd Syapul Othman
Mohd Yazreen Syahmie Yusoff
Mohd. Akmal Ali
Mohd. Akmal M. Kamel
Mohd. Amirul Airi Pauzi
Mohd. Arif Syakir Rosid
Mohd. Asfarizal Masmok
Mohd. Azhar Jusoh
Mohd. Azlan Harun
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Nur Najmi Basyeer Abdul Karim
Nur Shuhada Ariff
Nur Zafirah Abdul Hamid
Nurashiqin Sallih Udin
Nurfadhlina Chan Mahadie Chan
Nurhidayati Ahmad Sobri
Nurhuda Ibrahim
Nurul Aqilah Che Wan
Nurul Aziati Ab. Aziz
Nurul Farhanis Syatila Kamaruzaman
Nurul Najiha Sallehudin
Nurul Nassita Lias
Nurul Salma Adenan
Nusrah Alifah Ahmad
Ong See Ling
Pang Shek Qin
Radhuhaida Ramli
Rathi Sai Muniandy
Raziyani Ibrahim
Rohana Shapiin
Roshazia Aini Abu Bakar
Roshilawati Bidin
Rosmawati Ghazali
Rosnah Mah Hasan
Rozana Johari
Roza Mohd. Idris
Roziawati Mohd Razali
Saadiyah Ibrahim
Ummi Rodiah Abd Aziz
Santhana Dass A/L Irudaiam
Shamila Zainol
Siti Arbaiyah Bokhri
Siti Asiah Awang
Siti Hasshura Hashim
Siti Hawa Mohamad Ali
Siti Norasiah Darahman
Siti Norjannah Johari
Siti Qhairun Nisa Rahman
Siti Rokhaiya Biollah
Siti Sutina bt. Omar
Sufiah Hashim
Sugania Vijayan
Sumawati Amad Bugis
Suriati Muhamad
Syarifah Shafura Wan Abdul Molok
Vaani Muniandy
Wan Haifa Haryani Wan Omar
Wendy Ferrina Tarry
Wong Yien Ping
Yeo Moi Eim
Zawati Awang
Zeti Hafiza Zakaria
Zul Asyraf bin Mohd. Jamri
Zuraidah Roli
Ainul Yasmin Md Yusoff
Amatul Samahah Md. Ali
Amira Hanani Azali@Sazali
Amira Syahidah Nordin
Amirah Abdul
Arbania Ali
Christina John

Mohd. Fauzi Ismail	Chua Sing Ying
Mohd. Fauzi Salehon	Dayang Syahreeny Abang Mustafa
Mohd. Fazlan Abdul Ghani	Eleanor Daniella Lokman
Mohd. Fazli Long	Goh Ann Jie
Mohd. Husaini Rosli	Imelda Rantty
Mohd. JohariA. Latiff	Intan Ashikin
Mohd. Nasir Muhamad	Jumria Sutra
Mohd. Nazuri Rudin Jali	June Moh Hwei Yieng
Mohd. Nor Effendi bin Abd. Aziz	Kelly Teh
Mohd. Redhvan bin Arif	Kumari Geetha Muniandy
Mohd. Riduan bin Adnan	Laura Khor
Mohd. Rozhan bin Zakaria	Lim Mui Hua
Mohd. Sayuti bin Ismail	Lizawati bt. Mohamed
Mohd. Shafiq Abu Bakar	Marjorie Charam
Mohd. Sharifuddin Abdul Manan	Masazurah A. Rahim
Mohd. Shubli bin Md. Saad	Melinda Lau
Mohd. Shukri bin Yusof	Melissa Wong
Mohd. Sukri bin Jusoh	Noor Faizah Ismail
Mohd. Syafiq bin Manan	Noor Hanis Abu Halim
Mohd. Syairazi Che Man	Noorul Azliana Jamaludin
Mohd. Taufiq bin Md. Nordin	Noradiana Noran
Mohd. Yusof bin Mohd. Yasin	NorFatmahwati Yakup
Mohd. Zuraire bin Ab. Rani	Nur Amalin Nadia Mat Nasir
Muhaffiz Hamid	Nur Anissa Ainnina Daud
Muhamad Izzuan Razali	Nuraida Zulkifli
Muhamad Noor Zainal Abidin	Rabi Atun Abdullah (record not found)
Muhamad Nor Lamzahu Ismail	Rizmanisha Jasmin
Muhamad Yusof Nor Jasman	Rohaiza Asmini Yahya
Muhamad Zamri Abdul Murad	Sarmila Muthukrishnan
Muhammad Afiq Muhaimin Mohd Ashhar	Sarmiza Saperi
Muhammad Aliff Iqbal Abdul Hisam	Shaaleni Pandian Muruthy
Muhammad Amirul Razak	Sharifah Lia Farliana binti Wan Alias
Muhammad Amirul Siddiq Abd Rashid	Siti Naquiah Md Pauzi
Muhammad Anas Razali	Siti Nurul Annisa Temrin
Muhammad Asyraf Abd Latip	Sridevi Devadas
Muhammad Danial bin Abdullah	Stephenie Kawi
Muhammad Fadzil Haron	Syamsuria Abdul Latip
Muhammad Faizal Mohd	Umang Jabu
Muhammad Farouk Harman	Wen Chian Tan
Muhammad Fathullah Ruslan	Zarina Zainuddin
Muhammad Firdaus Ahmad Zamri	Nor Aida Suzana Abdul Rahman
Muhammad Hanaffi Ayob	Norazila Jelani
Muhammad Haziq	Nur Amalina Mohd Razikin
Muhammad Hazizi Ismail	Nur Simaa Ismail
Muhammad Helmi Mohamad	Nuramirah Abdul Manaf
Muhammad Izham Szali	Nurhidayati Ahmad Sobri
Muhammad Izzat Kamaruzaman	Nurul Nadwa Abdul Fatah
Muhammad Na'im Syafiq Othman	Meisam Tabatabaei
Muhammad Nur Ashry Bassari	Su Shiung Lam
Muhammad Safwan bin Othman	Yik Sung Yeong
Muhammad Safwan Khairul Asri	Rohani Mustafa
Muhammad Saufi Ahmad Khairuddin	Rosmaria Abu Darim
Muhammad Syafiq Izzuddin Abdul Hadi	Azizul Fariha Ghazali
Muhammad Thaqif Mohamad Bakri	Siti Nabila Mohd Sharif
Muhd. Ariff bin Abdullah	Hamidah Zainal
Muhd. Sharir bin Sha'arani	Fazlan Abu Bakar
Mazniza Othman	Nor Aida Suzana Abdul Rahman
Noor Azean	Intan Syazlin
Anuar Deraman	Haslizan Abdul Latif
YM Ku Kassim Ku Yaacob	Aiman Zulkiflee
Husaini Rosli	Mohd Zamri Mohamed Zin
Mohamad Hafiz Mushidi Shakori	Amirah Fatimah Md Nordin

Riduan Riduan
Mohamad N. Azra

Mohd. Hafizi Wan Hussin

Norway

Edgar Brun
Polyana Ferreira Da Silva

Karin Pittman

Phillipines

Lorenz Hans Reyes
Ma-ann Camarin
Mary Nia Santos
Jeryl Belle Rafanan

Joseph Carlo Vergel
John Paul Matthew Guzman
Sonia Somga

Republic of Korea

Olumide Olowe

Singapore

Jaglarz Anita
Markus Schrittwieser
Panqin Cai
Qinghui Tan
Siow Foong Chang
Zuridah Merican

Eng Yan Jie
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Poh Sze Choo
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Anuphap Prachumwat
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Jie Huang
Rungkarn Suebsing
Saengchan Senapin
Supraanee Chinabut
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Jiraporn Srisala
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Christopher Payne
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**THANK
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