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Zuridah Merican

Time to build alliances to improve productivity

Apart from human health, Covid-19 (coronavirus) poses a clear and present danger to the supply chain. When China shut down for business post Chinese New Year for more than 2 weeks, ships and containers were stuck at ports and the banking system took longer to normalise. In their Global Health & Crisis Response, McKinsey & Co's base case scenario predicted a global slowdown up to Q3 of 2020. So, what are the implications for the shrimp aquaculture industry?

In the past two years, both Ecuador and India have been gearing up to compete on the supply side. In 2019, India reported a production level of 800,000 tonnes and Ecuador at 600,000 tonnes. Both countries have targeted China which is estimated to have imported 650,000 tonnes of shrimp. While this supply exceeding demand scenario has been leading to a price war, Covid-19 and the supply chain disruption have further aggravated the situation. On February 18, Undercurrent News reported that Ecuadorian shrimp prices had dropped 15-20%.

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Does the shrimp industry have a crisis response? The initial responses have varied greatly from a more positive view in Ecuador to mixed signals in India and a pessimistic view from Indonesia and Vietnam. It is time to develop a business continuity plan and the three key words the shrimp industry needs to focus on are ***markets, margins and productivity*** and this is the theme for TARS 2020 to be held in Ho Chi Minh City, Vietnam.

The Asian intensive shrimp model has been beset by diseases, plunging average survival rates to 55% and increasing the costs of production. Margins have been squeezed as evidenced by some Indian farmers avoiding stocking during Q1 of 2019. The weak links in the production supply chain have already been identified; so the objective now is to find possible solutions.

The goal is to bring survival rates back to 80% and TARS 2020 will address these weak links. One weak link seems to be throwing SPF PL 12 into a pond water filled with pathogens and open to fluctuating weather conditions. Every industrial agribusiness, ranging from crops to poultry and livestock have addressed the challenges during the early stages of the life cycle and shrimp aquaculture should follow suit. We still want the grow-out stage in open systems. Therefore, adding climate controlled nurseries like RAS, with early stage nutrition and healthcare would provide optimal conditions for the fast growth genetics available today. Direct costs will increase but the upside from higher survival rates and economies of scale will reduce the cost of production per kg.

Unfortunately, the shrimp immune system is comparatively primitive and 'prevention is still better than cure' when it comes to disease mitigation. The critical challenge is that diseased shrimp do not feed hence the mode of delivery for any therapy is rendered impossible. This is where building immunity and functional feeds come in and yet functional feeds are going nowhere today. There are unanswered questions on what, when and how functional feeds work. There are indeed additional costs and it all boils down to minimising this and who bears it. It would be untenable for either the feed supplier or the farmer to bear it alone. This industry has worked in silos long enough but a crisis requires stakeholders to work together to create value along the production supply chain. The industry is solely dependent on farmers, i.e. without farming, all the support industries would automatically collapse. TARS 2020 proposes to work on building alliances between farmers and the hatcheries, feedmills and processing plants, respectively.

It is clearly noticeable that insurance companies avoid shrimp aquaculture. We have no risk mitigation plans and we have not learnt from past mistakes. Although the industry has accumulated sufficient data, there has been no one to analyse and make sense of it. With the emergence of start-up companies armed with artificial intelligence and internet-of-things, the industry today has the necessary tools to identify trends and determine trigger points. At TARS 2020, we encourage farmers to share their data so these start-up companies can develop risk mitigation programmes for the industry and insurance companies. This would ultimately make the industry more conducive for investment.

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Incredible India achieves record shrimp production at 804,000 tonnes in 2019

This announcement at the beginning of AQUA INDIA 2020, held from 31 January-1 February in Kochi, Kerala brings India as the world leader in farmed shrimp production to date. This was a surprise as back in October 2019, during GOAL2019, **Ravi Kumar Yellanki**, President of the Society of Aquaculture Professionals (SAP) had expected a dip in the 2019 production with a delay in stocking in the first half of 2019.

He attributed this 7.2% increase over that for 2018 (750,000 tonnes) to a changing landscape in shrimp farming in India. "Another record is the total exports figure of 668,000 tonnes. We have never achieved this volume before. In fact, in each month, from August to December, we were exporting roughly 60,000 tonnes and in December, it was 63,000 tonnes. How different is this? Previously, we did most of the exports in June-July."

India also cemented its role as a leading exporter of farmed shrimp; into the US at 40% of total shrimp imports in the US. India's share in the Chinese market was at 27% in 2019. Participants at this Kochi meeting demonstrated their pride on this achievement in shrimp exports as MPEDA Chairman Shri K S Srinivas announced during his keynote address. The Marine Products Export Development Authority (MPEDA) is the agency responsible for the export promotion of seafood products including its overall development. (see pages 13-16).

While the reported CAGR for global farmed shrimp production was around 3.5% from 2006-2012 and 6.2% from 2015-2018, the highest growth was in 2014 at 20% (Andersen et al., 2019). India's CAGR for farmed shrimp production was 21.26% for 2011 to 2019 (MPEDA, 2020). "Fisheries and aquaculture contribute 5% to agriculture GDP and we are drawing attention. With the government's Blue Revolution, we now have INR 7,500 crores (USD10 million) Fisheries and Aquaculture Investment Development Fund," said Ravi Kumar.

State of shrimp farming in India

Ravi Kumar described some paradigm shifts in 2019 which had contributed to the additional production. "Our stocking season usually starts at the end of January or early February and for some farms, there will be a second or third crop. In 2019, except for Andhra Pradesh, farms in other states delayed stocking. Usually during the last five months of the year, production is lowest but in 2019 production during this latter part of the year was record breaking. We attributed the good performance in the last five months of 2019 to more conducive weather conditions with relatively lower temperatures. Better prices since June 2019 also encouraged stocking."

According to Willem van der Pijl, Seafood Trade Intelligence Portal, Indian farm gate shrimp prices have remained relatively flat throughout 2019 but the fact that prices did not dip too much towards summer and in fact slightly improved, encouraged farmers to stock their ponds for a new crop.

"There are some positives." said Ravi Kumar, "Production increases came from Andhra Pradesh, Odisha and West Bengal, mainly from inland farms with 0-5ppt salinity. In a recent survey conducted by SAP in collaboration with the Aquaculture Pathology Laboratory, University of Arizona, USA, we found that at low salinity, susceptibility to *Enterocytozoon hepatopenaei* or EHP, our major threat in 2019, is reduced. Therefore, I advocate a change in stocking pattern and avoid the hot summer months when the mortality rate due to white faeces syndrome (WFS) and EHP is high."

Some participants commented that partial harvesting allowed farms to benefit from market demand for a range of sizes, from small size shrimp 100/kg to size 30/kg. In Andhra Pradesh and Tamil Nadu, 4 crops/year was possible for 100 count shrimp with 70 days/crop. Ravi Kumar added that staggered harvesting of many sizes, prevented a glut in harvest and gave entry to various markets.

The sector has been preparing for higher production over several years. Its shrimp feed production capacity is around 2.5 million tonnes per year and 2019 production was 1.2-1.3 tonnes. Feed conversion ratio (FCR) has been improving, from the 1.74 in 2016 to 1.5 in 2019, attributed by lower density stocking and early harvests. The 600 hatcheries have a total capacity of 125 billion post larvae but production was only 71 billion post larvae in 2019. Recent expansion in processing capacity can handle 1 million tonnes/year. The potential land area is 1,150,000ha.

Early indications are a better first crop and possibly a higher production in 2020, perhaps at 900,000 tonnes. However, Ravi Kumar asked, "Can the market absorb the extra volumes? In 2019, we had a good market penetration. The Chinese market is important for India, but we already have a new challenge. We consider the demand for WSSV-free frozen shrimp, a 'trade barrier' which requires intervention of our government. Nevertheless, what is important is to create our own domestic demand, initially move to a shrimp consumption of 100g to 200g/capita/year."

Ravi Kumar Yellanki (third right) with participants and SAP executive members; Dr Victor Suresh, General Secretary (third left) and S. Muthukaruppan, Adviser (centre).

Related article: The changing face of shrimp aquaculture in India at AquaIndia 2020, (see pages 8-12).

Merger creates largest fully integrated barramundi enterprise

In January, Singapore's Barramundi Asia Pte Ltd and Allegro Aqua Pte Ltd, announced a merger to combine resources and catalyse knowledge-based, sustainable practices and create one of the largest fully integrated barramundi enterprise with a global footprint.

This is a merger of advanced genetic selection, vaccines, sustainable grow-out practices, downstream processing and global market reach integrated into one concerted value chain. Anchored in Singapore, both companies are committed to contribute to the '30 by 30' food security roadmap and would continue to supply vaccinated superior quality Asian sea bass *Lates calcarifer* or barramundi fry to local farms to help boost production. It integrates the success of R&D programmes by both companies mainly on the selection of superior traits using genomics and marker-assisted selection and autogenous vaccines. It comes on the back of Barramundi Asia's success in scaling up production and market distribution of its barramundi products globally.

Autogenous vaccines and John's Sea Bass® fry and fingerlings

Barramundi Asia's fingerlings are already vaccinated by proprietary autogenous vaccines produced at its subsidiary UVAXX which also provides health monitoring support. Combined with Allegro Aqua's elite St John's Sea Bass® fry and fingerlings, production volumes will be enhanced. Allegro Aqua will continue to work with UVAXX and the Temasek Life Sciences Laboratory (TLL) to develop future generations of vaccinated barramundi fingerlings for Barramundi Asia's nucleus of regional farms; as well as other Asian seabass farms in Singapore and Southeast Asia.

Barramundi Asia (BA), founded in 2008, operates the largest barramundi ocean farms in Singapore and Australia; off the Southern Isles of Singapore and in the Buccaneer Archipelago, Australia. It is in the midst of developing 6,600ha at Brunei's Nankivell Offshore Aquaculture site. It also operates its own recirculating aquaculture system (RAS) hatchery and nursery.

Allegro Aqua (AA) is a start-up established in 2018 by scientists from Temasek Life Sciences Laboratory (TLL) and backed by Temasek Life Sciences Accelerator (TLA) to commercialise the fast growth and higher survival strain of the Asian sea bass, known as the St John's Sea Bass®. This is the result of 15 years R&D by TLL and AVA (now known as the Singapore Food Agency-SFA) at the Marine Aquaculture Centre on St John's Island (see page 26-27). The start-up also contracts partners to grow out these fingerlings using strict protocols. TLA is a Singapore-based biotech and agritech incubator with seed funding provided by the Lifesciences Innovation Fund (TLIF) and various other angel investors.

Vaccinating fingerlings. Barramundi Asia's subsidiary UVAXX produces proprietary autogenous vaccines and also provides health monitoring support. Photo credit: Barramundi Asia

Barramundi Asia markets its barramundi under the trade name Kühlbarra®. It is the only farm with a BAP (Best Aquaculture Practices) certification in Singapore. Photo credit: Barramundi Asia

Andreas von Scholten, CEO at Barramundi Asia said, "Allegro Aqua's expertise in life sciences research and innovation will empower Barramundi Asia to realise synergies from improved genetics. With this merger, our quest is to make barramundi the "Salmon of the Tropics". In 2015, Barramundi Asia pioneered a full "Farm-to-Fork" value chain in Singapore, serving B2B and B2C customers. Its barramundi under the trade name Kühlbarra® is endorsed by multiple Michelin-rated chefs, celebrity restaurateurs and premier hotel chains. It is the only farm with BAP (Best Aquaculture Practices) certification in Singapore.

Andrew Kwan, Director at Barramundi Asia added, "What will be truly industry transforming is the plan to supply superior vaccinated barramundi fingerlings to other farmers in Singapore, coupled with a guaranteed buy-back scheme. Farmers opting for this will follow a strict prescribed feeding programme and adhere to the international standards of husbandry practices adopted by Barramundi Asia because it will be a branded product."

Koh Soo Keong, Chairman at Allegro Aqua said, "As an investor in Allegro Aqua and mentor under TLA's Entrepreneur-in-Residence programme, I am delighted to be able to personally witness and help see to fruition a vision that TLL and AVA started more than 10 years ago. I continue to believe that with the commitment and passion of the staff of both companies, as well as the R&D support by TLL, we will hear more successes in the coming months."

Peter Chia, CEO, Allegro Aqua and TLA said, "This merger reflects our belief that Singapore has the ability to put together and build a robust aquaculture ecosystem, with innovation, trans-disciplinary capabilities and global market access being important pillars of that vision. I am very fortunate to have a very dedicated crew and hope that this merger will spur the development of a unique life sciences development nexus that will help build resilience and enhance Singapore's and Asia's food security and nutritional needs over the next few years." www.allegroaqua.com/ www.barramundi.asia

Do not underestimate the resilience capacity of the Chinese market

In a press release dated 11 February 2020, the National Chamber of Aquaculture of Ecuador (Cámara Nacional de Acuacultura-CNA) said it expects that at the end of the holidays in China, domestic trade will begin to show signs of recovery reactivating the demand for shrimp very soon. It added that the announcement of Chinese authorities regarding the completion of the long holiday generates a positive expectation for the Ecuadorian shrimp industry. CNA expects a fast recovery of the demand especially that focusing on safe food, said the Executive President, José Antonio Camposano. This is positive news because Ecuador is recognised for its excellent reputation as one of the largest shrimp producers worldwide with a clean record in traceability. "What happened in China will undoubtedly generate more attention from consumers concerning the conditions of the food they buy. With Ecuadorian shrimp, Chinese consumers can have the certainty that they are buying not only the best shrimp in the world but the safest, the healthiest and the only one with reliable traceability and the most natural one."

He added that Ecuador was a pioneer country in obtaining the Aquaculture Stewardship Council (ASC) certification which is one of the most demanding in the world in terms of health and social responsibility. Likewise, the Ecuadorian industry has added a considerable number of certifications which has made it worthy of an excellent position in the international market, including the Chinese market to which it sells 67% of its production.

"Ecuador not only complies with the most demanding standards and certifications in terms of health, safety, conservation and social responsibility, but also has the only shrimp certified as free of antibiotics through the 'Sustainable Shrimp Partnership' initiative which has the endorsement of the ASC, the World Wildlife Fund (WWF) and the Sustainable Trade Initiative (IDH)," said Camposano. "We are focusing all efforts to guarantee Chinese consumers 100% of the traceability of our product, taking care of every detail in the production chain, so that they can resume consumption levels." In 2019. Ecuador exported USD 2 billion of shrimp to China equivalent to 422,727 tonnes.

Vannamei shrimp farming pilot trials in Bangladesh

The government has decided in principle to introduce vannamei shrimp farming in Bangladesh. It has allowed Shushilan, a non-government organisation in Khulna and Agri Business Enterprise in Chattogram to launch two separate one-year pilot projects on vannamei shrimp farming under the supervision of the Department of Fisheries and the Bangladesh Fish Research Institute, according to www.newagebd.net in January. This move follows demands from industry for more shrimp raw material for processing plants and as the black tiger shrimp was losing its market due to high production cost. For several years, the government was reluctant to allow vannamei shrimp farming as green activists claimed that its culture would be harmful for biosecurity. Md Golam Mostafa, former president of the Bangladesh Frozen Foods Exporters Association, appreciated the government's decision but said that the government should come up with the infrastructure facility for the culture of the species.

Agreement for aquaculture disease prevention

In March, Pebble Labs and Virbac announced the signing of a comprehensive commercial agreement for aquaculture disease prevention. Based in Los Alamos, New Mexico, USA, Pebble Labs is a biotechnology company developing solutions that address some of the world's biggest challenges in agriculture and aquaculture with the mission of improving global food security. Virbac (VIRP) is the world's seventh largest veterinarian pharmaceutical group and a leading animal health player in aquaculture. The companies will focus on validating and commercialising a solution for white spot syndrome virus (WSSV) in farmed shrimp, a pathogen that can cause 50% crop loss and has been estimated to reach up to USD3.5 billlion each year.

Currently the aquaculture industry relies heavily on probiotics, synthetic chemicals and antibiotics to fight the diseases which can be devastating to shrimp farms worldwide. The Pebble Labs Directed Biotics™ technology harnesses an animal's natural immunity with a bacteria and redirects it to suppress WSSV. "Pebble Labs has the first sustainable technology to safely and effectively address the viral pathogens we are facing in large-scale aquaculture," said Pierre Henning, DVM, Director of Aquaculture Division, Virbac. "Working with Pebble Labs to share their solution with farmers is a high priority for our aquaculture division this year, and we are committed to moving this project along quickly."

Terms of the joint development agreement include executing feasibility studies to validate Pebble Labs technology, solution development plans, regulatory clearance and approval and discussion of future products and commercial applications in addition to treating WSSV in aquaculture. "Pebble Labs is looking forward to seeing Directed Biotics technology in the pond this year," said David G. Morgan, President, Pebble Labs. "This agreement with Virbac moves our revolutionary products toward the market which will reduce the need for antibiotics in food production and improve food security worldwide." www.pebblelabs.com

Investments for international expansion in insect production

In June 2019, Protix, the global leader in insect production opened its new innovative insect factory in Bergen op Zoom with an investment of €40 million. In March, it has attracted investor Rabo Corporate Investments as shareholder. Protix wants to use the capital from Rabobank's investment franchise to scale up the production of insects in the Netherlands and accelerate the roll-out of its international expansion. Protix breeds larvae from the black soldier fly and processes them into sustainable ingredients like proteins and lipids.

"We want to reinforce our international leadership position in this new industry," said Kees Aarts, founder and CEO of Protix. "A 'Global Technology with Local Impact' aligns with our vision to strive for a food system in balance with nature worldwide." Joost Vogels, Investment Manager of Rabo Corporate Investments said, "Rabobank Group has been involved with Protix since its founding more than 10 years ago. We are proud that they have become a global leader in insect production and Rabo Corporate Investments looks forward to support them in their international expansion plans." www.protix.eu

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The changing face of shrimp aquaculture in India

The biennial meeting of stakeholders addressed current and future challenges in marketing shrimp and overcoming diseases, mainly EHP.

Notwithstanding being a leader in farmed shrimp production, the industry in India recognises that it must keep up with changes vis-à-vis the global trends in shrimp aquaculture and markets and address evolving threats. This was the goal at the biennial AQUAINDIA 2020 held in Kochi, Kerala from 31 January to 1 February, which was attended by 370 participants. Over the years, since it started in 2005, Aqua India, organised by the Society of Aquaculture Professionals (SAP), has developed into a leading regional conference showcasing industry trends. With international attention, it is also a gateway to get to know Indian aquaculture.

The programme was organised along the theme, "The changing face of Indian aquaculture". It comprised sessions on marketing shrimp with reference to global trends in demand and supply, shrimp diseases, and future solutions. There was a session on diversification options, to farm finfish and domesticated *Penaeus indicus*, which reflected the national call to add more species to India's seafood exports and not be dominated by the vannamei shrimp.

Some recent developments were raised by **Ravi Kumar Yellanki**, SAP President. State-wise, shrimp production in 2019 was led by Andhra Pradesh which showed an increase of 13.9% from 2018 to 570,000 tonnes in 2019. Gujarat showed a 20% drop in production to 45,000 tonnes, mainly because of the monsoons. Higher production in Andhra Pradesh, Odisha and West Bengal came mainly from inland farms, which produce vannamei shrimp in 0-5ppt salinity. (Figure 1)

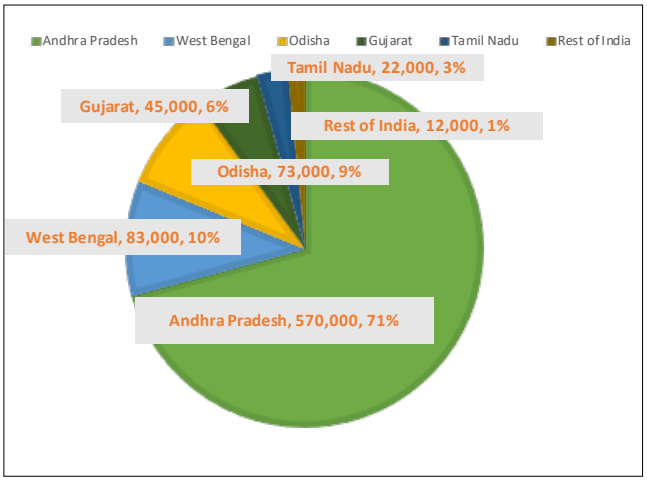

Figure 1. Statewise contribution to shrimp production in India (tonnes and percentage) in 2019 (Ravi Kumar Yellanki, 2020)

The total shrimp broodstock import was 228,800 pieces which was 10% lower than in 2018 and post larvae production was 71 billion against 73.6 billion in 2018. This means a higher post larvae efficiency at 11.30 (tonnes of shrimp/million post larvae) was achieved in 2019 compared to 2018 (10.2).

India's position in global shrimp markets

Willem van der Pijl, representing Seafood Trade Intelligence/ Shrimp Tails, Netherlands used custom data, which he believes is much more reliable than production data to discuss "Supply trends in global shrimp markets". He added that the only drawback is that domestic consumption is not included in the custom data. He described the monthly supply trends and leading product forms in 2019 by leading shrimp exporting countries, including India.

Willem van der Pijl, John Sackton (centre), and Elias Sait, Secretary General of the Seafood Exporters Association of India (SEAI)

"In the first four months of 2019, Indonesian shrimp exports registered a weak trend, just like India. In summer, exports were better and prices improved to USD5/kg for size 60/kg in June. By October, Indonesia's exports were 154,801 tonnes, just 1% behind the total for 2018. Vietnam was 6% ahead of its 2018 performance. Prices rose from July (USD4/kg, size 60/kg) to USD5.5/kg, size 60/kg) towards the end of the year. Vietnam exported more cooked shrimp, which were preferred by EU markets. Ecuador, closed the year with a growth rate of 25% at 634,000 tonnes of exports whereas prices were USD4/kg for size 60/kg."

His analysis showed that India exported 85-90% vannamei in its shrimp basket. "Indian farmers were clearly encouraged by good weather conditions and favourable prices. During summer, shrimp harvests were strong and from August onwards shrimp exports easily outperformed the weaker end of 2018. India exported less headless shell on shrimp (HLSO) but increased raw peeled shrimp."

The US is the major market for all three Asian countries, ranging from 23.5% for Vietnam, 68% for Indonesia and 54.7% for India. China is the major export market for Ecuador at 54.2%." Each country has their unique mix of product forms: Ecuador with 75% HOSO exported to China, India with almost 70% raw frozen block shrimp, and Thailand peeled and finished products. To the US, Indonesia supplied mid and large sized shrimp; India is the leader with HLSO, mainly size 15-20/kg and Ecuador small size HLSO.

John Sackton, Founder Seafood News.com/Seafood Datasearch, USA raised a question in his presentation "Global demand for shrimp in 2020": How can the market absorb all these growth? India and Ecuador will have 20.7% and 16.37%, respectively of the global shrimp trade in 2020.

Sackton provided estimates on volumes imported by major markets up to November 2019: USA, 858,000 tonnes; Europe, 905,000 tonnes and China, 624,000 tonnes. In terms of growth over 2014-2018, imports grew by 4%/year in the US; 59.1%/year in China and 1.4%/year in EU markets. Japan's demand decreased by 0.6%/year. Globally, demand went down by 7.4%. The EU and Japan will not be able to absorb significantly higher volumes of shrimp and volumes will be at the levels during the past two years.

"China is upending world shrimp markets. Up to the third quarter of 2019, it imported 217,358 tonnes of shrimp from Ecuador and 107,632 tonnes from India. It is also beginning to diversify where it gets its shrimp. It surpassed the US with imports in the third quarter with more than 150,000 tonnes of head on shrimp (converted). Imports were driven by falling domestic production and increased growth came from the expansion of frozen shrimp distribution into mid-tier cities, expansion of cold chain distribution and delivery options, supermarket and online shopping."

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Drivers for growth in 2020

Both van der Pijl and Sackton gave their take on what to expect in 2020. Van der Pijl said that more shrimp production is expected from new farms such as those in Venezuela and Oman, more inland farms in Andhra Pradesh or new farms in Gujarat. More vannamei will come from the transition from monodon shrimp to vannamei shrimp in Bangladesh, Sri Lanka and Vietnam's Camau region.

"Sri Lanka may contribute 50,000 tonnes in 3 years. Intensification will play a crucial role in the further growth of farmed shrimp production such as from 15PL/m^2 to 30PL/m^2 in Ecuador, or from 70PL/m^2 to >150PL/m^2 in Vietnam. Shortening of production cycles will increase the volume per year," said van der Pijl.

Sackton said, "Growth in global shrimp markets will come from China and the US. US shrimp consumption has increased to 4.4lbs or 2 kg/capita/year and shrimp import volumes have set records for the past 2 years. But prices in China have dropped; Ecuadorian HOSO shrimp was just above USD3.40/lb (USD7.48/kg, size 30-40/kg) in early January 2020 from above USD3.5/lb (USD7.7/kg) in early 2019.

In the US, peeled shrimp is driving market growth as well as larger sizes (up to 26-30 HLSO). This fits into India's exports of processed and peeled large shrimp; the former grew 54% and the latter 44%, as compared to 2018. Growth was simultaneous with the fall in price, but lower prices were passed to consumers which in turn supported more growth. Stability in prices also encouraged promotions.

Sackton said, "Price is the most important factor driving current production growth. While Ecuador may have the ability to increase to 850,000 tonnes, they must sell HOSO, because they do not have the plant capacity for peeling or processing. If China reduces demand/price for Ecuadorian shrimp, the only option for Ecuador is to accept a lower price or reduce stocking. India can reach one million tonnes of production. Sale of processed shrimp to China is increasingly important."

"In 2020, without the growth in China and US, to absorb the product, pricing will be much lower. The new coronavirus is a wild card as it causes a contraction in economic activity. China recently stopped imports of live shrimp," added Sackton.

Can certification help sell more shrimp? In response, **Greg Brown**, Global Aquaculture Alliance (GAA) which has the Best Aquaculture Practices (BAP) certification said, "Buyers want assurances that the products from their supply chains consistently meet the guidelines, specifications or expectations. They require that their suppliers achieve third-party certification according to international standards." He noted that with multiple standards in the market, there are financial costs, audit fatigue and confusion as to which is the "best" certification among them. Therefore, in April 2013, the Aquaculture Stewardship Council (ASC), BAP and GlobalGAP joined forces to offer a joint certification. This reduces duplication of effort for processing plants, farms, hatcheries and feed mills that undertake certification by more than one organisation.

Dealing with diseases

In his introduction to this session, moderator **Dr D Ramraj**, Past President of SAP and President of AISHA (All India Shrimp Hatchery Association), said that reactions to diseases varied in 2019. Some farmers postponed stocking to March and April which was also linked to low prices, and some reduced stocking density to counter diseases related to water quality. The productivity range was large, ranging from 2.5-2.7 tonnes/ha/crop to as high as 5-10 tonnes/ha/crop. One issue is that many small holders share the same creeks and sometimes implementing ideal biosecurity concepts may not work.

Shrinibas Mohanty (second left) with his Avanti Feeds team. On the left is Vinny Madhuri, Alltech India and on the right, Vilas Autade, Cargill Animal Nutrition, India.

"Disease threats in shrimp farms are numerous," said **Dr Amerneni Ravikumar**, Partner at Alpha Biologicals, India. Diseases encountered in broodstock include vibriosis and filamentous bacteria, while in larval and post larval rearing, farmers encountered the zoea-2 syndrome, mysis mold syndrome, vibriosis with hepatopancreas tubule constrictions and spirochaetes. *Enterocytozoon hepatopenaei* (EHP) was reported from the nursery to grow-out stages. During grow-out, diseases such as white spot syndrome virus (WSSV), infectious hypodermal and hematopoietic necrosis (IHHNV), infectious myonecrosis virus (IMNV), white faeces syndrome (WFS), slow growth, running mortality syndrome (RMS), loose shell, black spot and eyeball degeneration, have been reported.

To combat diseases, **Dr KV Rajendran**, Central Institute of Fisheries Education, India said, "The way we deal with diseases in shrimp aquaculture requires a fundamental change from reactive to proactive. There is a need for a pathobiome concept and industry has to play a larger role." In managing disease, Rajendran made several suggestions, among them, the re-implementation of conventional microbiological and microscopic protocols, which have been practised previously, to test the health of post larvae instead of the absolute reliance on PCRs. Adding a nursery is another layer of biosecurity as well as a management tool to increase growth of shrimp and shorten production cycles. "Cluster farming and better management practices (BMP) adoption are ways for small-scale farmers to minimise disease impact. In 2019, there are 10,175 farmers managing 10,728ha of shrimp farms."

Dr Marcela Salazar, Benchmark Genetics discussed the genetics tools available today. "From the point of view of accuracy, mass selection is the best and we can use the immune priming of shrimp, but biosecurity is the risk. Family selection gives 50% accuracy while in genomic selection, it will depend on the accuracy of the markers selected." She also discussed crossing of WSSV susceptible and resistance lines. Genotyping showed a 10% gain in WSSV resistance per population. Epigenetics can be used to control disease. Studies with artemia showed that non-lethal heat shocks can induce epigenetic inheritance of phenotypes across three successive generations.

Together with the Aquaculture Pathology Laboratory, University of Arizona, USA, SAP conducted a survey on the prevalence of EHP outbreaks and on its co-occurrence with WFS. **Dr Luis Fernando Aranguren Caro** reported on these findings as well as other experiences with EHP and WFS from a survey in Indonesia and Latin America. In India, the survey covered 16 farms in Andhra Pradesh, Gujarat and Tamil Nadu where 15 out of these 16 farms were infected with EHP. Is WFS a predictor of EHP endemic regions? asked Aranguren Caro who described the evolution of pathogenesis of WFS in EHP endemic areas. He indicated a strong association between WFS and EHP, in EHP endemic areas. EHP in combination with other enteric pathogens including septic hepatopancreatic necrosis (SHPN) and other unknown factors can cause WFS. Results also indicated that EHP was highly associated

with high salinity. In ponds with low salinity (3-5ppt), the EHP load was low.

Farming challenges and solutions

The moderator of this session, **Dr Manoj Sharma**, Mayank Aquaculture, Gujarat, said that for every problem and challenge, there are solutions. "There are lessons to learn on what works and what does not work in shrimp farming in India," explained **Shrinibas Mohanty**, Avanti Feeds, India. "My mantra is to harness each other's core competencies."

Some examples were on changing practices during the hot summer months when diseases such as RMS proliferate. Mohanty suggested to reduce feed by 15%/day and not feeding in the afternoon and to increase aeration to 1HP per 300kg biomass to counter poor water quality. With high bacterial loads in ponds, the trend is to use a central drainage system but channelling the effluents into shared creeks is counterproductive. "In Thailand, sludge treatment is compulsory before discharge. They have a Whats App group to self regulate such as when to discharge; the Department of Fisheries alerts farmers using a flag system."

With temperature, water salinity and climate changes, advocated following some of the disruptive practices in different regions in India. "In summer, Gujarati farmers reduced stocking density by 30% to 20-30 PL/m^2 and had better crops and good feed conversion ratio (FCRs). WFS occurs with high temperatures and reducing effluents load in creeks and taking care of the environment is crucial. In two villages, Kavali in Nellore and Cheyyeru in East Godavari, farmers took crop holidays to avoid disease and adverse weather conditions. Subsequently, survival rate increased to 80-90% from the previous 30-40%."

Other suggestions included using larger PL20, ensuring farm hygiene to avoid contamination of containers with spores and with on-farm fermentation, to use freshwater or sterile pond water to prevent an over proliferation of the harmful *Vibrio* bacteria. He commented, "There has been a proliferation of nurseries in India; in 2019 the number was 242. The larger post larvae from nurseries are definitely better for crop success. But once infected with EHP, the disease will stay. Although survival rates in nurseries averaged 71.5%, in nursery-based grow-out farms, survivals averaged only 51.5% in 2019. Therefore, we need to support farmers with technical knowhow and farm management."

In shrimp nutrition, **Dr Alex Obach** from Skretting's Aquaculture Research Centre described several new developments such as image analysis and quantitative histology which show morphological changes induced by different feeds. Knowing what happens in the hepatopancreas at the cellular level, such as haemocytes infiltration is crucial as the organ is involved in disease, health and digestibility. A software to measure shrimp behaviour can help in developing new attractants. Optimising feeding is important too; this can be achieved by precision feeding with the use of sensors to measure real time water quality parameters and automatic feeders which can also be remotely controlled.

As disease is a major threat, Obach said, "As an industry, we should be focussing on prevention and a prophylactic approach via functional feeds. This must be holistic and should include tools like genetics, biosecurity etc. Obach outlined steps in developing functional feeds but added that these will only work hand in hand with preventative measures.

Two alternative farming technologies were discussed. An intensive indoor shrimp farming system in Singapore developed by **Dr Farshad Shishehchian**, Blue Aqua International, Singapore and aquamimicry as a viable and sustainable way to produce shrimp

Veerasun Prayotamornkul (second left) and SAP's executive committe members, Manoj Sharma (third left), Dr Akshaya Panigrahi (third right) and Dr Saji Chacko, Vice President (right).

by **Veerasun Prayotamornkul**, Aquamimicry Technology and Thai Organic Shrimp Farming Group.

The indoor farms uses green water recirculation systems (RAS) in AgroDome™ to bring down air temperatures by 7°C. At a stocking density of 1,300 PL/m^2, the production can reach 15 tonnes/1,000m^2. The unique pure oxygen distribution system was also described. Veerasun said that aquamimicry is going back to nature, by combining natural estuarine conditions with natural zooplankton blooms, supplemental nutrition and synbiotics to suppress growth of pathogenic bacteria, reduce water exchanges and produce single cell protein *in situ*. Veerasun explained that a large population of copepods appears after broadcasting of the fermented rice bran paste and stocking takes place 2-3 weeks after. In his system, he prefers stocking 0.1-0.2g juveniles at 50 PL/m^2. Shrimp are harvested at size 30-40/kg.

Next issue: Diversification options in India with farming of the Asian seabass and its nutrition, farming the murrel and domestication of *Penaeus indicus*.

It is not just about exporting seafood

Although export volumes are up, MPEDA needs to address challenges in ensuring quality seafood from India, prepare for market changes and for the future, with diversification of aquaculture, says Chairman K S Srinivas.

Year 2019 has been notably significant for the Indian seafood export sector. It made its mark with USD 6.8 billion of total seafood exports. In 2019, India's share of imported shrimp in the US was 40 % with a volume of 282,584 tonnes. India had already replaced Thailand in 2013 as the leading shrimp exporter to the US. In 2019, it shipped USD one billion of shrimp exports to China. It is the second largest exporter of shrimp to the European Union (EU) and of frozen shrimp to Japan.

It is not just statistics and managing exports that will sustain India's position but also addressing and overcoming internal challenges; mainly in vannamei shrimp farming and especially if India wants the image of a sustainable producer of this export commodity, said Chairman **Shri K S Srinivas IAS** of the Marine Products Export Development Authority (MPEDA). To maintain India's lead in seafood production, he has outlined several approaches: increasing productivity, species diversification, bringing aquaculture to inland areas and revival of the monodon shrimp. Although aquaculture grew at an impressive 800% in the last decade, only 15% of available area is being utilised. The processing segment has 32,297 tonnes/day capacity but is only 30% utilised.

For Srinivas, coming from the Indian civil service to lead MPEDA in 2018, getting to know the multi-faceted seafood and aquaculture business continues to be a learning curve, yet exciting. He wants to introduce global standards to aquaculture in India. One accolade that he is proud of, is the registration of more than 70,000 farms with geotagging. In this interview at MPEDA's headquarters in Kochi, together with his team, Srinivas described his work and ambitions for the sector during his tenure as chairman.

MPEDA is a statutory body under the Ministry of Commerce & Industry, India, and was set up in 1972. It is the agency responsible for the export promotion of marine products including its overall development. It has a regulatory role in quality control throughout the supply chain, registration of exporters, processing plants, cold storages, pre-processing and handling centres etc and enrols hatcheries and aquaculture farms for traceability. In aquaculture, it is responsible for R&D for commercial aquaculture, development of technologies for aquaculture production, best management practices and certification. The Rajiv Gandhi Centre for Aquaculture (RGCA) is the R&D arm of MPEDA.

MPEDA Chairman Shri K S Srinivas is very excited over the progress with the e-enrolment and geotagging of farms programme which was started in 2013-2014.

Achievements, markets and aquaculture

According to Srinivas, "In 2019, our volumes of seafood production and exports have increased. But there was a slight decrease in terms of value to USD6.8 million, attributed to lower prices and to the depreciation of the INR. However, in INR terms, the amount was marginally higher than in 2018. We exported nearly 14 million tonnes of various seafood; shrimp from capture and farming totalled 41% and out of this 90% was vannamei shrimp. In terms of value, shrimp accounted for 69%." During the opening of Aqua India 2020, Srinivas also announced that from April 2019 to November 2019, the shrimp export volume was 313,617 tonnes.

The question is whether India's shrimp aquaculture industry can keep growing. Srinivas said there is no doubt that India can expand shrimp production, but it needs to be less dependent on imports of shrimp broodstock. This is a reason why MPEDA has called for proposals to set up Nucleus Breeding Centres (NBC). "RGCA is playing an important role in aquaculture development with supply of seedstock and this shows that MPEDA not only promotes seafood products from India but is also pushing for aquaculture development."

A subsidiary of MPEDA is the Network for Fish Quality Management and Sustainable Fishing (NETFISH). It is encouraging domestic consumption of seafood. Nightly, this food truck sells various seafood produced by local processors.

Value added seafood products on display at the MPEDA headquarters in Kochi. India has 442 tonnes/day capacity for value adding processing.

Diversification in aquaculture

The vannamei shrimp is the leading commodity in India's seafood export market. While Srinivas is promoting increasing productivity in shrimp farming, at the same time he advocates for diversification of aquaculture in the long term. "Our productivity is an issue. We have in Gujarat, a productivity of 7-8 tonnes/ha, in Kerala only 1 tonne/ha and in Andhra Pradesh, 5-6 tonnes/ha. Can we increase this to an average of 8-10 tonnes/ha nationwide?

The diversification plan includes the seabass, pompano and tilapia. It is also to revive the farming of the monodon shrimp. RGCA's Multispecies Aquaculture Complex (MAC) in Kochi is supplying fingerlings to spearhead farming of these species (see pages 20-25).

"The target species for export are the seabass and tilapia, which are now being farmed for local markets. Farmers sell tilapia at INR200/kg (USD 2.8/kg). Unless we have the critical mass, we can't start exporting. The tilapia market is very competitive, and we need to be concerned about low prices. But tilapia is farmed in freshwater and grows fast," he said.

Today RGCA is doing the R&D but a single institution cannot meet the needs of this vast country. "We cannot only focus on the coastal states, we want to go to the inland states and use ponds and reservoirs. We can expand aquaculture into inland states such as Haryana, Punjab, Telangana etc. Therefore, we really need the private sector to come in," said Srinivas. MPEDA is willing to transfer technology and provide training, which it is already doing for the state employees of the Fisheries Departments in Tamil Nadu, Kerala, Andhra Pradesh and Maharashtra. "We are ready to commercialise, we want at least one hatchery in each state to act as the engine for growth. I invite foreign entrepreneurs too, in fact in aquaculture 100% foreign direct investments are encouraged."

Caution on antibiotic residues in shrimp exports to the EU and US

In 2018, the number of rejections by the European Commission (EC) for shrimp imports into the EU was 37 with 13 attributed to antibiotic residues. Some processors have been delisted from exporting to the EU. The US FDA has issued alerts on detection of antibiotic residues in shrimp imports from India. B. Sreekumar, MPEDA Secretary, updated that after continuous campaigns to create awareness, the incidences went down: in 2019, EC issued

> "Once we have a bad image, it will not be easy to remove that image. Therefore, we work towards not allowing for any rejections." - K S Srinivas

11 alerts, out of which 4 were due to antibiotic residues. The US FDA rejected 30 containers and 6 were because of detections of antibiotic residues.

Srinivas expressed his concern. "This is a serious issue which we, as the authority responsible for exports, need to address. It is not an easy task as we have so many small farmers, some with farms less than 2 acres (0.8ha) and they number around 100,000. The farmers say that they are not using antibiotics directly. We suspect positive detections in samples could be through use of feeds produced by other industries or from top dressing of feeds."

The objective is to ensure the quality of Indian products in international markets, "If we do not give quality products, buyers will reject our shrimp. Once we have a bad image, it will not be easy to remove that image. Therefore, we work towards not allowing for any rejections."

Assuring quality of shrimp products

India has 563 processing plants and 347 are approved to export to countries in the EU. MPEDA has quality control laboratories in Kochi, Bhimavaram, Nellore and Bhubaneswar with facilities to test seafood samples for heavy metals, dyes, pesticides and antibiotic residues using advanced technology. Nationwide, it has 11 laboratories with ELISA analysers to ensure only antibiotic residue free farmed shrimp are exported.

MPEDA imposes a pre-harvest test (PHT) where samples from farms are tested for the presence of antibiotic residues. "The EC has made this compulsory for exports into the EU. Other countries have not asked for such tests. Processors exporting to the EU countries are also asking us to issue a certificate of health," said Srinivas.

NRCP

Since 1998, there is also a nationwide national residue control programme or NRCP. MPEDA staff take samples from aquaculture farms, processing plants, hatcheries and feedmills to test for residues from antibacterial/veterinary medicinal products and environmental contaminants. When there are residues, MPEDA will warn them and write to the Export Inspection Agency (EIC), the competent authority for exports and to the state governments. MPEDA cannot take any action against hatcheries/farms as it does

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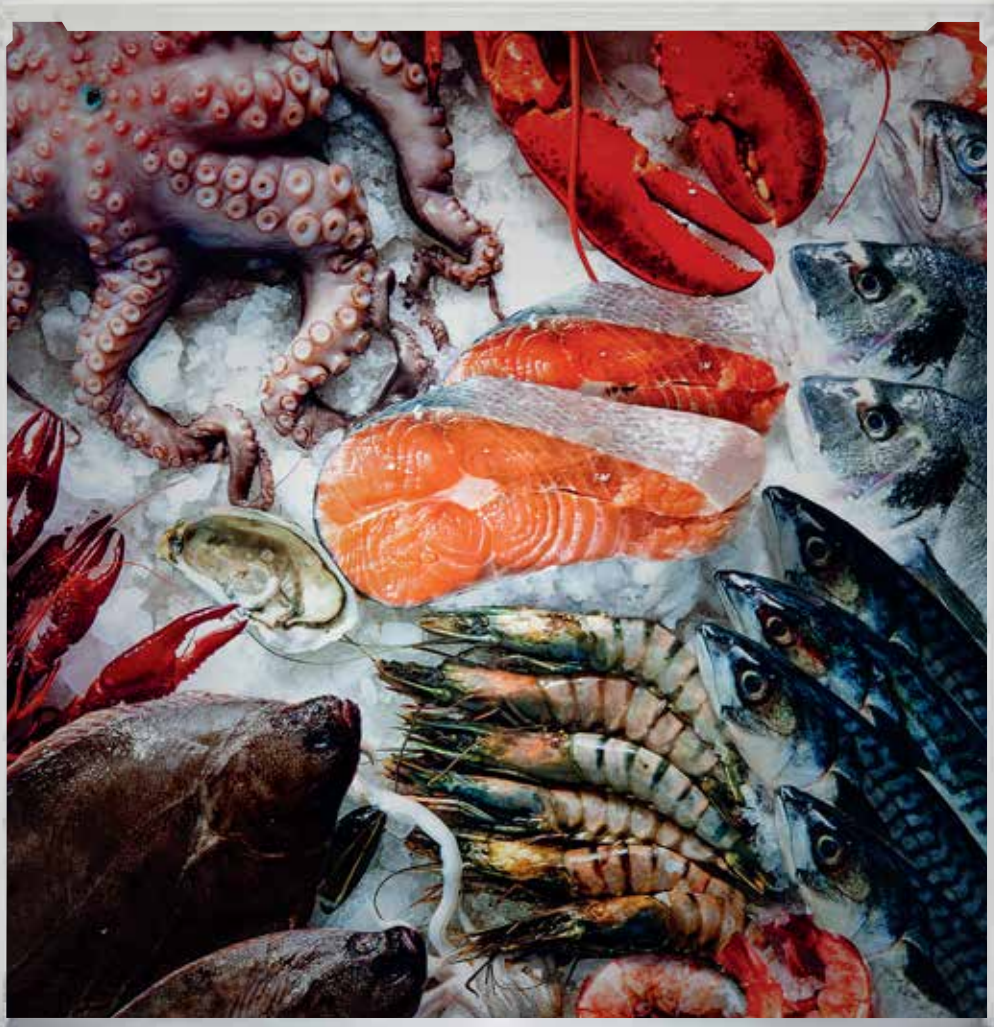

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not have the power to do so. The EC is monitoring this programme and makes annual visits; the next visit is in March/April.

Sreekumar described the progress under the NRCP. "In 2016, we tested 4,816 samples and 112 samples were positive. In 2019, only 15 samples were positive for antibiotic residues. This confirmed the level of awareness is increasing and efforts to put a stop to antibiotics usage is working. From our point of view, we are moving very fast. Each year, we conduct more than 100 campaigns, and we are strict; if we find any hatchery with antibiotic residues, we call the hatchery up and the farms using the post larvae from the hatchery."

Enrolment cards for the first mile

This relates to the strategy to know the supply chain well. The first mile connectivity is the weakest point in India's seafood industry. It is complicated as there are many agencies involved in regulating the aquaculture industry; for instance, the Coastal Aquaculture Authority (CAA) regulates farms 2 km from the coast and has registered 11,000 shrimp farms. State governments are responsible for inland farms. There are about 450 shrimp hatcheries.

Chairman K S Srinivas (seated) and his team from left, Dr S Kandan, Project Director, RGCA; K Sivarajan, Deputy Director; Dr T G Manojkumar, Project Manager, MAC-RGCA, Kochi and B Sreekumar, MPEDA Secretary

"This means that for any requests for traceability, it can be done all the way along the chain from the end port to the farm. All these are available online. I believe that this is a first in the global shrimp industry. It is costly but worth it." – K S Srinivas

Srinivas explained that after taking note of all the constraints, MPEDA decided to initiate the enrolment programme. As of 30 January, this year, it has registered 71,409 farms engaged in export-oriented aquaculture, covering a water surface area of 178,000ha. These are in Andhra Pradesh, Tamil Nadu, Odisha, West Bengal, Gujarat, Maharashtra, Karnataka and Kerala. This means that for any requests for traceability, it can be done all the way along the supply chain from the end port to the farm. The information is available online. "I believe that this is a first in the global shrimp industry," said Srinivas. "It is costly, but worth it."

The enrolment is mandatory to accompany the pre-harvest test certificate (PHT) for exports to the EU countries. (See box). There are 95 hatcheries yet to be enrolled; this will be completed by the first half of 2020.

Proposed certification programme

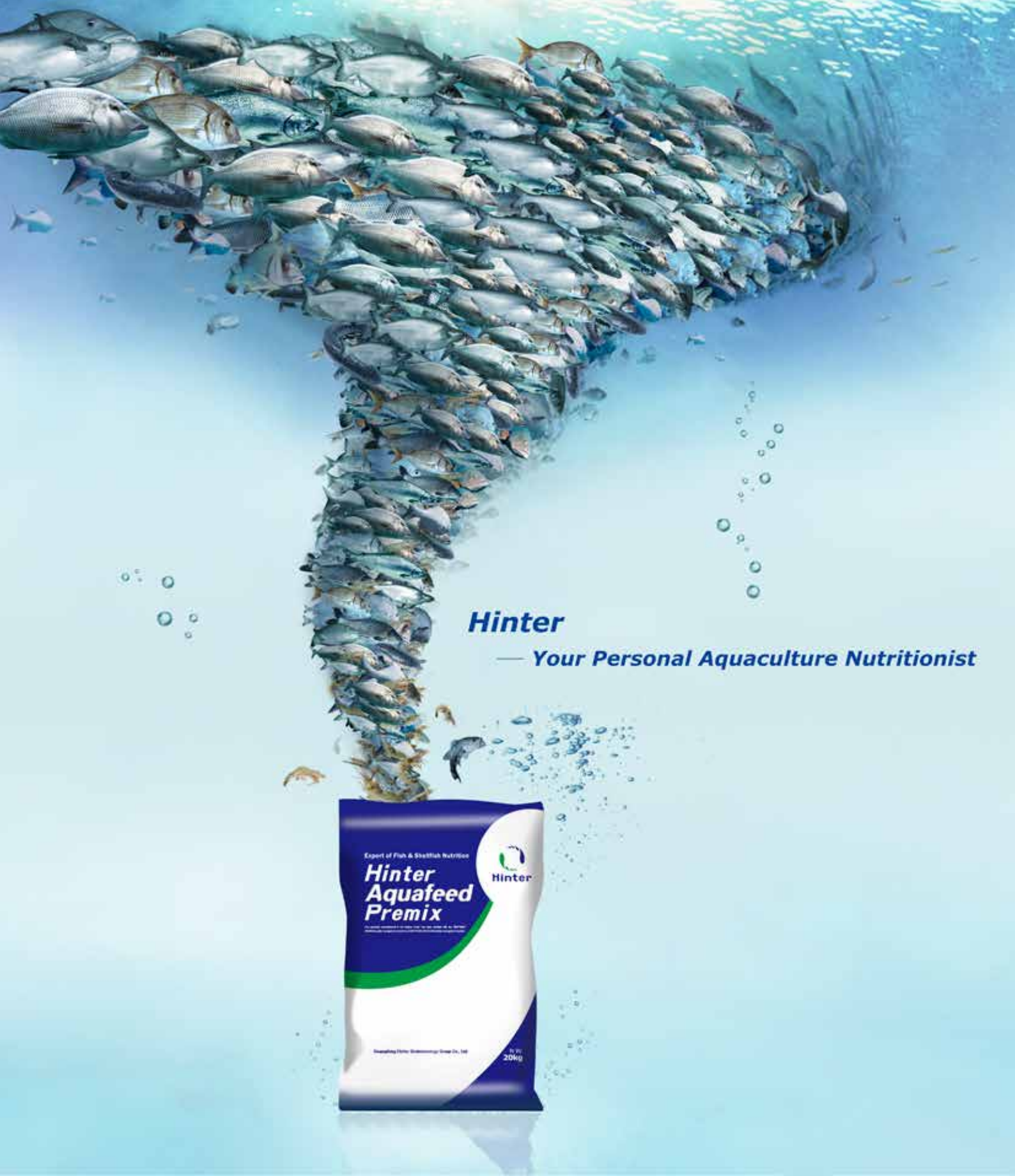
During his opening address at GOAL2019 and recently during Aqua India 2020, Srinivas announced that MPEDA will have a certification programme for India's shrimp farmers. Acknowledging that there are several certification programmes world-wide, he explained that most farmers are unable to take up certifications like Best Aquaculture Practices (BAP), Aquaculture Stewardship Council (ASC) etc as these are expensive for them. "Our certification will be much cheaper and will be one tenth the price."

E-enrolment and geotagging farms

Started in 2013-14, this is an enrolment of farms. An individual, group or corporate body can apply for this enrolment. Throughout various stages, local staff will visit the farm for geotagging by taking waypoints using GPS devices. The farm area and coordinates will be uploaded on real-time google maps. The farms get a unique code which allows for tracking from pond to harvest, to purchaser, to processor, to the port in India and finally the port in the importing country.

The aim is to enroll all freshwater/brackish water export-oriented aquaculture farms and aquaculture hatcheries to ensure traceability and quality maintenance of the NRCP, PHT and disease surveillances. It gives the census of aquaculture farms all over India, facilitates proper monitoring and extension of assistance. The system as shown on the right, provides with one click on google maps, the location and other details.

The enrolment is free of charge for the grow-out segment. There is a separate scheme for the hatchery segment where there are fees, between INR 5,000 (USD 68) and INR 10,000 (USD 139) for fish and shrimp hatcheries, depending on production capacity. An inspection committee will visit the hatchery and may make suggestions for modifications wherever required, prior to approval.



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Understanding Artemia

Biology, characteristics and processing technologies

When you consider that global Artemia cyst consumption has multiplied 30,000 times (from a modest 100kg worldwide in the seventies to well over 3,000 tonnes today), you will understand that the Artemia market for commercial aquaculture is constantly under pressure – on the buyer as well as the supplier side. For aquaculturists to efficiently and sustainably source, buy and use Artemia, a thorough understanding of its fascinating biology is needed.

Artemia biology: a natural enigma

The first thing to keep in mind is that Artemia or brine shrimp is a live organism, specifically a small crustacean that has developed a unique defence mechanism to survive in harsh conditions. More particularly, Artemia has a unique reproductive system. When conditions are favourable, the species is live-bearing. But during the hardship of winter, increase in salinity or other extreme environmental conditions, the brine shrimp change reproductive strategy and starts producing dormant encysted embryos capable of withstanding severe dehydration. This enables the population to escape extinction. The embryos develop up to the gastrula stage, a point where they reach about 4,000 cells when the development is arrested by greatly reducing the metabolism of the embryo to to a level which is close to zero (biologists call this 'diapause'). This is the stage at which they can be harvested, processed, conditioned and stored as dormant or quiescent live organisms, ready to be used as feed in fish and shellfish hatcheries.

The cysts

The cyst's shell that encapsulates the brine shrimp embryo is a natural marvel that up to this day intrigues biologists worldwide. Artemia themselves secrete a rigid noncellular shell to cope with the extreme environmental stresses. The complex noncellular cyst shell consists of two main regions: the outer region, secreted by the shell gland, and the inner region formed by blastoderm cells comprising the embryonic cuticle. They form a delicate, but very effective membrane that allows water, oxygen and CO_2 to pass through but shields the embryo from the uptake or leaching of bigger molecules and harmful external influences such as UV light. Safely embedded in this breathing shell, the embryo can sit out its diapause and wait for the ideal conditions to resume further development and hatch.

Hatching mechanism

When outside conditions improve to the point where it is safe for the brine shrimp larvae to come out of their shell, the embryos will respond to a number of triggers and resume their metabolism. They will start to metabolise and convert their carbohydrate reserves into glycogen (as energy source) and glycerol. The latter hygroscopic compound will make the cysts take up more water, eventually increasing the osmotic pressure inside the cyst up to the point that the shell bursts. Now the nauplius, still enclosed in the hatching membrane (also called umbrella stage) is released. Within the hatching membrane one can see the nauplius moving its appendages. Within the next hours, the hatching enzyme is secreted in the head region of the nauplius and the free-swimming instar I nauplius is released. It is important to note that glycerol is a good bacterial substrate, in particular for *Vibrio* species.

Several elements play a crucial role in this delicate process that aquaculture professionals need to imitate to optimally hatch Artemia cysts:

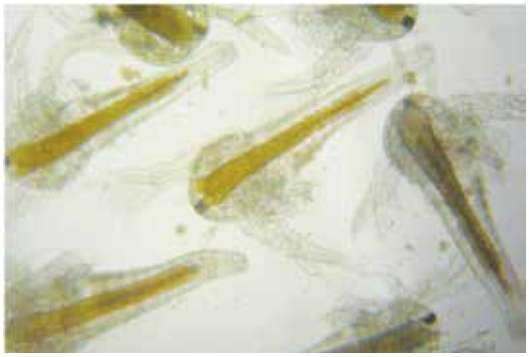
- Temperature: the optimal water temperature for Artemia to hatch out is around 29°C±1°C.
- Salinity: although cysts can hatch at salinities as low as 5g/L (when the lowest levels of glycerol have to be built up to reach the breaking stage, and thus more energy is left in the nauplius), most hatcheries use regular seawater for hatching Artemia cysts. The optimal salinity for most of cysts is approximately 25g/L.
- pH: the final step in the hatching process (the release of the free-swimming nauplius) is made possible by a hatching enzyme that has a maximal activity in the pH range of 8 to 8.5.
- Oxygen: Artemia cysts can hatch at oxygen concentrations as low as 1mg/L, however, for optimal hatching, oxygen levels above 4ppm are recommended.
- Light: light of sufficient intensity is the trigger to initiate the hatching metabolism as soon as all other conditions are favourable. Depending on the type of cyst, a continuous illumination between 2,000 and 4,000 lux is needed.
- Cyst density: although cysts can be hatched at much higher densities, most hatcheries do not apply more than 2-3g/L. This avoids the cysts shading their own light and allows to maintain high oxygen levels without inducing foaming or mechanical injury of the hatched nauplii.

The nauplii

About 18-24 hours after metabolism has resumed, a free-swimming Artemia larva appears (Instar I nauplius). Within a few hours, the Instar 1 nauplius moults into a second larval stage (Instar II nauplius). From then onwards, it takes about another 11 consecutive moults in about 10 days for the Instar II nauplius to develop into an adult brine shrimp. For aquaculture hatcheries, the first two larval stages are important.

The Instar I stage does not have a functional digestive system; whereas in Instar II, the mouth and anus are open and the nauplius becomes an active filter feeder that takes up particles from less than 1µm up to about 20µm. Instar II can be enriched with specific products to enhance its nutritional quality.

In most crustacean hatcheries, Artemia are fed directly after hatching at the Instar I stage when the nauplii still contain a maximal energy content. But it also means that, if they are left in the hatching tank, they consume their own energy reserves and will have lost up to 20% of their energetic content when moulting into the Instar II stage.



In fish hatcheries, Artemia is usually fed at Instar II after being enriched. Some super quality Artemia that are very rich in highly unsaturated fatty acids (HUFAs) can also be fed at the Instar I stage. This is especially done during the first larval days when the mouths of the fish larvae are too small to ingest the larger Instar II nauplii.

As soon as possible, however, the growing fish larvae are fed with enriched Instar II nauplii. This stage has a fully developed and functional digestive tract and can effectively consume exogenous feed of appropriate size. This property of active ingestion is exploited to load (enrich) the Instar II nauplii with nutritional products. Fish larvae that feed on these enriched Instar II nauplii not only benefit from the Artemia itself, but also, and probably more importantly, from the ingested nutritional components. This method has proven to lead to better survival rates, growth, robustness and stress resistance.

Efficient use of Artemia: technology to the rescue

Given the growth of aquaculture and the fact that global Artemia resources are being exploited to their full sustainable capacity, a highly efficient application on the hatchery floor is an important challenge for the future. Even the best quality cysts on the market remain highly sensitive to storage and hatching conditions. Besides this, a meticulous application of feeding protocols and the adequate separation of the nauplii from their empty shells are crucial.

To make sure that hatcheries make the best possible use of the available Artemia (i.e. to get a maximum number of nauplii out of the cysts they purchase), new technologies are continuously developed to eliminate the risk factors in Artemia hatching. INVE Aquaculture has recently updated its Artemia product portfolio to offer three added-value technologies.

The patented SEP-*Art* technology applies a coating on the cysts which responds strongly to magnetic fields to flawlessly separate the nauplii from their shells. SMArt, another new INVE Aquaculture technology, allows cysts to hatch optimally despite sub-optimal illumination conditions. The new D-Fense technology uses plant extracts to suppress the growth of bacteria such as *Vibrio* sp during the Artemia hatching process.

Looking ahead, INVE Aquaculture's R&D department confirms that future strategies will include the elimination of other risk factors such as the incubation temperature of Artemia cysts.

A new era has started for Artemia, moving from simple raw material into a complex product that is more and more adapted to the customers' needs.

Diversification into finfish aquaculture in India

MAC is a business model for fingerling supply to push farming of alternative species in ponds and reservoirs across India and cages in the backwaters of Kerala

By Zuridah Merican

The Marine Products Export Development Authority (MPEDA) aspires to have other products in its seafood basket, aside from the vannamei shrimp. It targets a seafood export revenue of USD17.8 billion by 2030, from the current USD6.8 billion achieved in 2019. Diversification into finfish production is the role of its R&D arm, the Rajiv Gandhi Centre for Aquaculture (RGCA).

RGCA's USD 1.1 million Multispecies Aquaculture Complex (MAC), located in Kochi, Kerala has supplied fingerlings and post larvae to 2,034 farmers since it started production in December 2018. Spread over 3.6ha, there are 6 nursery ponds and enclosed indoor nursery tanks for the rearing of fry to fingerlings of the Asian seabass, pompano and tilapia. To produce monodon shrimp post larvae, there is a quarantine and hatchery complex. (See article in the next issue of AQUA CULTURE Asia Pacific) Picture credit: RGCA

Diversification has been long in the plan and preparation. Since 1999, RGCA has embarked on the breeding of the Asian seabass *Lates calcarifer* at its facility in Thoduvai, Nagapattinam, Tamil Nadu. It has also demonstrated the farming technology in Karaikal, Puducherry in 2000 and at Mahendrapalli, Tamil Nadu in 2012. There are hatcheries and demonstration farms for the cobia *Rachycentron canadum*, pompano *Trachinotus blochii* and *snapper Lutjanus argentimaculatus*. Then, there is the collaboration with the WorldFish Center on the selective breeding of the Genetically Improved Farmed Tilapia (GIFT) *Oreochromis niloticus* in Manikonda Village, Andhra Pradesh, since 2008.

"Our next step is to push for more farming of these species. This will require a steady supply of fingerlings for cages-in-ponds and cage culture in open waters and reservoirs. This is the aim of the Multispecies Aquaculture Complex (MAC), an INR8 crores (USD1.1 million) facility in 9 acres (3.6ha) of land on Vallarpadam Island, Kochi, Kerala. In December 2018, we began to produce fingerlings of all the species mentioned above, except for the cobia," said Dr S. Kandan, Project Director, RGCA.

Dr S Kandan, Project Director, RGCA (right) and Dr TG Manojkumar, MAC Project Manager

From wetlands to multispecies aquaculture complex

Back in 2002, this site housed a hatchery which trained many technicians in monodon shrimp post larvae production. In 2017, MPEDA's then chairman Dr A. Jayathilak IAS, posed the question to the team on whether this site can be redeveloped. "It was a tall order, as this area is below sea level and was often inundated," said Kandan. "The design and construction were very challenging; we had to raise the land. But RGCA's dedicated team took up the challenge and developed a multispecies aquaculture complex, the first of its kind, with a hatchery for the monodon shrimp and a hi-tech nursery for the year-round supply of the fingerlings of several finfish. In the end, we managed to complete the work within a year. We started on 13 September 2017 and under the proficient leadership of the present chairman Shri K. S. Srinivas, IAS, the facility was completed on 24 August 2018. On December 17, 2018, we achieved our first production. To date, we have provided seedstock to 2,034 farmers."

With some expansion from its original design, MAC has six nursery ponds, with sizes ranging from 600-650m^2 and a monodon shrimp quarantine and hatchery complex for high health post larvae production. The location allowed for nursery grow-out of both marine species (pompano and Asian seabass) and of the freshwater tilapia. A new activity here is the breeding of the freshwater/brackish water pearlspot fish, *Etroplus suratensis*, the official fish of Kerala. The popular fish is retailed at INR600/kg (USD8.3/kg) for 300g fish.

Fast tracking finfish farming

India has a vast coastline, and as such it should be farming more marine fish species, especially since the Government of India (GOI) will soon issue guidelines on the leasing of open waters. Kandan said, "Currently, most of the marine fish farming is over here in the west coast of India. On the east coast, the Bay of Bengal is too rough for marine fish farming and furthermore, there are at least 2-3 cyclones in a year."

In Kerala, the state government has a blue revolution plan and they are already putting out cages in the backwaters of Kerala. "The backwaters are a blessing for Kerala. Here, conditions are ideal for cage culture in protected waters. From November to June,

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Nursery ponds for tilapia and pompano, with hapa nets (right) and feeding trays (left)

salinity of the backwaters begins to increase from 20ppt. By end of January, it rises to 30ppt. These are ideal conditions for the farming of the cobia over 6 months as the marketable size is 3-4kg only. Then when the salinity goes down to 0-15ppt it is the time to farm the tilapia."

In the backwaters of Kerala, farmers stock square and round cages. Trials are being conducted to use larger round cages. In cages-in-pond systems, initially farmers stock the cages and when the fish are larger, they release the entire stock into the pond.

Asian seabass and pompano fingerling production

RGCA is pushing for the farming of three marine fish species: the Asian seabass, pompano and cobia. MAC obtains the Asian seabass fry (1-1.5cm) from the hatchery in Thoduvai. In 2019, the hatchery produced 2.8 million fry. Over at MAC, fry is reared on locally produced extruded feeds to two sizes: over 30 days to 3.5cm fingerlings for pond culture and over 45 days to reach 4-5cm fingerlings for direct stocking in cages. Some farmers demand larger fingerlings of 8-10cm.

Project (year started)	Production capacity/ year
Asian seabass seed production, Thoduvai, Nagapattinam, Tamil Nadu (1999)	3 million
Mangrove mud crab seed production, Thoduvai, Nagapattinam, Tamil Nadu (2004)	1 million
Aquaculture demonstration project Asian seabass, Karaikal, Puducherry (2000) Aquaculture demonstration project Asian seabass, Mahendrapalli, Tamil Nadu (2012)	30 tonnes 250,000 crablets
Tilapia (GIFT) hatchery and seed production, Manikonda, Andhra Pradesh (2008)	6 million
Marine finfish seed production project, Pozhiyoor, Kerala (2009)	Pompano/cobia – 1 million
Multispecies Aquaculture Complex (MAC), Kochi (2018)	20 million disease-free monodon shrimp post larvae 3 million GIFT tilapia fingerlings 0.5 million Asian seabass fingerlings

Table 1 RGCA's R&D in seed production and farm technology demonstration of finfish and crustacean aquaculture (extracted from Kandan, 2020)

> "Our strategy is to make the fingerlings readily available to get farmers interested in marine fish farming and then create a demand for fingerlings. After seeing our nursery, we want the private sector to start a nursery culture business as well."
> – S Kandan

The demand of Asian seabass fingerlings is in two sizes, 3.5cm fingerlings for pond culture and over 4-5 fingerlings for direct stockings in cages

Using a PVC pipe to provide feed directly into the feeding tray via gravity.

GIFT tilapia juveniles

Male fry of GIFT tilapia (1-1.5cm) which have been treated with methyltestosterone are transported from the RGCA hatchery in Andhra Pradesh to MAC for grow-out into fingerlings of three size ranges (3-4cm, 4-6cm and 6-7cm) for stocking in freshwater ponds. Tilapia fingerlings are sold at INR 6.8 to INR 10 each or USD 0.94-1.38 each, based on size.

The hatchery production is 6 million/year. "We have an agreement with WorldFish in Penang, Malaysia to carry out some genetic selection of the GIFT tilapia supplied by the latter. To date, the 7th generation at the RGCA's hatchery has 8% faster growth over that of the 6th generation. Recently, the collaboration was extended for another 3 years," said Kandan.

Dr T G Manojkumar, MAC Project Manager explained that by June, salinity in the backwaters will be around 15ppt and in July, it will be down to 0ppt and this is when farmers will stock tilapia which is tolerant to salinity up to 15ppt. "Tilapia cultured in 15ppt waters has better flesh texture and tastes better than tilapia farmed in freshwater. When cultured in freshwater, farmers will keep the tilapia in 15 ppt water for 2-3 days to remove the muddy taste and improve flesh quality."

Manojkumar added, "Fish consumption is high at 22kg/capita/year in Kerala as compared to the national average of 8kg/capita/year. The demand for tilapia is especially high in Kerala during the 15 June to 15 August period when there is a ban on trawling in marine and brackish waters. For the Keralites, fish is essential in the daily diet."

In 2019, 0.5 million juveniles were produced at the complex. Asian seabass fingerlings of 5cm are sold at INR 24/fingerling (US 33 cents/fingerling). There is an additional INR 2(US 0.03 cents) for every 0.5cm length increase. Farmers can produce Asian seabass of 1kg size over a grow-out duration of 8-10 months. Fish are sold as fresh fish in local markets at INR 500/kg (USD 6.92/kg)

The hatchery in the south, some 5 hours away, in Pozhiyoor, Kerala carries out breeding of the pompano, cobia and red snapper. While the nursery rearing for cobia fingerlings is continued in this hatchery, pompano fry is brought over for nursery rearing at MAC to grow to 4-5cm fingerlings. Pompano fingerlings of 5cm are sold at INR 10 each (USD 1.38 each). There is an additional INR 2 (US 0.03 cents) for every 1cm length increase.

"Our strategy is to make the fingerlings readily available to get farmers interested in marine fish farming and then create a demand for fingerlings. After seeing our nursery, we want the private sector to start a nursery business as well. Without a steady supply of fingerlings, cage culture in Kerala and in the rest of India will not be successful.

"Among the marine species, the Asian seabass is the best candidate for farming," said Kandan during his presentation at Aqua India 2020. "It has a delicate flavour and can be farmed at 0-35ppt salinity. The fish is fed on extruded feeds with 45-50% crude protein or on trash fish. The final survival rate is 97% over the 145-day production cycle during which there are several stages of fish grading."

The demand is for tilapia fingerlings of three size ranges (3-4cm, 4-6cm and 6-7cm) for stocking into cages in the backwaters or in ponds. Picture credit: RGCA

Kerala also has several large reservoirs. Kandan said that tilapia fingerlings have been given to women self-help groups, since it is a relatively easy fish to farm and requires feeding only twice daily. After 5 months, fish can be harvested at 0.5kg from a stocking size of 4cm. Extruded and pelleted feeds are available from local feed millers.

"Before we began to supply tilapia fingerlings to farmers, around 2-3 million spurious tilapia fry/fingerlings were imported and mislabelled as GIFT tilapia. I believe this was the reason for the spread of tilapia lake virus (TiLV) in our reservoirs. This forced us to screen all our broodstock in the hatchery," said Kandan.

Since 2018, MAC has supplied 3 million fingerlings to the farmers in Kerala. However, the demand is around 6 million fingerlings. "It is important that we continue to supply the farmers here in Kerala or else they would either import or transport fingerlings over 48 hours from the hatchery in Andhra Pradesh. We are extending technical consultancy to the state government in Kerala for developing two GIFT hatcheries. There are interests from five companies to start tilapia hatcheries and two from the state government of Tamil Nadu.

Currently, production is just enough for the local fresh and chilled fish markets. But looking ahead, MPEDA expects production volumes, mainly of the tilapia and Asian seabass, to be high enough for export. "The future is diversification, away from relying only on the vannamei shrimp," said Kandan.

Kandan is very proud that MAC is self-sustaining. It has 42 employees comprising of staff holding post graduate and graduate degrees. All operating costs are covered by the revenue from sales of fingerlings and post larvae. "We are providing a service to farmers and yet it is a self- sustaining business model."

"Today, we do not have a precise target on production. We only want to get the big entrepreneurs to get out of the shrimp syndrome and look at other options such as the tilapia," said Kandan. "There are already nine entrepreneurs interested to set up hatcheries: 3 in GIFT tilapia, 3 in Asian seabass, 2 in mud crab and 1 for multispecies finfish such as the cobia and pompano." At the hatchery in Puducherry, RGCA has developed the breeding and rearing of crablets and the grow-out of the mud crab *Scylla serrata*. The National Fisheries Development Board (NFDB) is ready to assist with financing of up to 25% of the project costs.

Although RGCA and MAC are ready to transfer technology as well as help in the start-up of new ventures in marine fish hatchery, nursery or grow-out activities, Kandan noted that working with the marine finfish such as the Asian seabass requires passion and financial commitment.

GIFT broodstock. The tilapia breeding is at the hatchery in Manikonda, Andhra Pradesh. Picture credit: RGCA

The MAC team comprises 42 technical and non-technical staff.

One of the projects by MPEDA was the introduction of aquaponics in the Cherai village, Kerala together with the Pallipuram Service Co-operative Bank (PSCB). Since 2016, Cherai has the unique distinction of being India's first aquaponics village. MPEDA supplied fish seeds, feeds, water quality detection kits and technical training and the bank gave guidance and financial support to interested farmers. Picture shows a model for visitors. (https://www.thebetterindia.com/167334/kerala-farmers-aquaponics-diy-how-to-cherai/)

Spearheading research in marine aquaculture in the region

Fast growing Asian seabass has reached commercialisation and RAS can be used to control hatchery environments to prevent big belly disease.

The Singapore Food Agency (SFA)'s Marine Aquaculture Centre (MAC), located on St John's Island in the open southern waters of Singapore was established in 2003. The aim of the centre is to expand on R&D in aquaculture genetics, nutrition and health in marine aquaculture. SFA was formed as a new statutory board under the Ministry of the Environment and Water Resources on 1 April 2019. The SFA brings together food-related functions carried out by the former Agri-Food & Veterinary Authority of Singapore, the National Environment Agency and the Health Sciences Authority.

MAC has a footprint of about 1.8 ha. Lim Huan Sein, Director of SFA's Aquaculture Department, who was one of the early proponents of MAC, said that MAC was first set up for development of large-scale hatchery technology and related R&D areas. Today, there are a total of 14 blocks in MAC, including purpose-built research facilities, incubation modules and other auxiliary facilities such as the seawater intake pump house. The R&D on tropical marine aquaculture at MAC is carried out in collaboration with local and international institutes of higher learning, research institutes and industry partners. Besides R&D, MAC provides technical support to local farms on sustainable farming practices and technology, hosts learning journeys for schools and organisations, conducts practical sessions and mentors interns from polytechnics and universities.

Fast growing and higher disease resistance Asian seabass

Since its setup, MAC has focused on genetics and broodstock development of the Asian seabass (*Lates calcarifer*), and the development of large-scale hatchery production technology. During the tour of MAC, Liang Bing, Senior Scientist at MAC, described the progress on the selective breeding programme on Asian sea bass using marker assisted selection (MAS). This has been a major focus at MAC, as seabass is a major culture species and the use of faster-growing seabass can improve the productivity of local farms. MAS, which began in 2003 in collaboration with Temasek Life Sciences Laboratory (TLL), has enabled them to make inroads in the genetic development of the seabass. MAS has enabled more accurate selection of commercially important traits and maintained genetic diversity in selection stock.

"While the focus on the F1 generation was growth, we have worked on other traits in addition to growth for the F2 and F3 generations, such as omega-3 content in the flesh and disease resistance. MAS has allowed us to predict the performance of offspring with higher accuracy and shorten selection time. We started with a wide diversity of stocks, from Singapore, and the region (Indonesia, Malaysia and Thailand). We genotype and tag the fish individually with a microchip, which can be identified by a handheld scanner. DNA markers enables traceability of fish offspring to their parent stocks. The superior fish are selected not only based on their phenotype such as the size and body weight, but also their genetic information such as family tree. Each trait is not determined by single genes but a group of Quantitative Trait Loci (QTLs)."

On the progress to date, Liang Bing said," We have established genetically superior seabass lines with increased growth rate

Tanks holding seabass at the Marine Aquaculture Centre on St John's Island, Singapore.

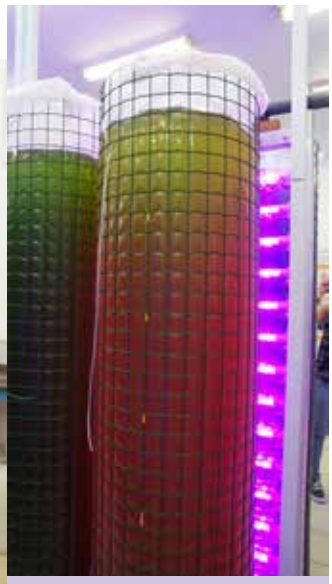
of more than 30%. Mass crossing of 15 males with 15 females is carried out through induced spawning which usually involves more fish compared to natural spawning. This is more suitable for the selective breeding programme because there would be more genetic combinations in the offspring. We have different tanks for specific purposes, i.e., for fast breeding and egg production for commercial sales. The pure breeding lines are for producing and selection of the next generation while composite lines are for commercial production. Setting up a composite line can protect intellectual property (IP) when the eggs are sent to the farms and it can also generate hybrid vigour for better performance."

In June 2018, Allegro Aqua Pte Ltd, a start up by TLL scientists began to scale up commercial production of the superior breed of Asian seabass, now branded as St John's Seabass®, after this island (see page 5).

By using indoor recirculation aquaculture systems technology (RAS), MAC has achieved a survival rate of 40-50% from hatching (DPH0) to weaning of seabass fry at DPH30. RAS can also be used to control culture environments. For example, the bacterial big belly disease, common in seabass farming, can be controlled by reducing salinity from 30ppt to 10ppt over the susceptible period of DPH20-DPH40. Salinity of >28ppt is necessary for early larval rearing and the key is the gradual lowering of salinity without stressing the larvae and to maintain 10ppt by day 30. As seabass is euryhaline, seabass fry growth is not affected at lower salinity and further lowering the salinity to 5ppt can suppress disease manifestation if there is any clinical sign of big belly disease.

Five year-old, 4-8 kg seabass for breeding of F4 generation

Xu Qunying holding a pack of concentrated algae, Nannochloropsis oculata and describing microalgae production at MAC



Mass production of the microalgae Nannochloropsis oculata at MAC. This microalgae is the feed for rotifers and artemia, which are the live feed for newly hatch seabass larvae.

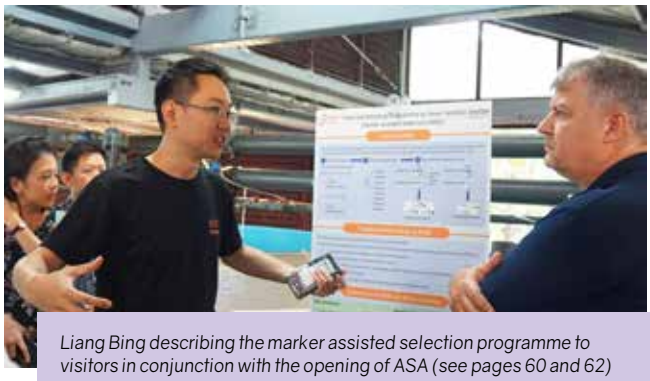
Microalgae technology

MAC maintains pure lines of several microalgae as feed for the fish larvae, including *Tetraselmis suecica*, *Chaetoceros* sp, *Isochrysis galbana* and *Nannochloropsis oculata*. It produces and supplies concentrated live microalgae to local hatcheries and farms. MAC also conducts research on other live feeds for aquaculture such as rotifers and Artemia. Rotifers and Artemia constitute a major part of the diet for marine fish larvae during early stages of fish rearing and the production of live feeds is essential in any marine fish hatchery. "The focus at MAC is to use RAS to have a continuous culture of rotifers *Brachionus* spp. In this way, we can enhance stability and prolong culture period at higher rotifer density. Furthermore, we can reduce the labour required and make rotifer production more cost-effective," said Xu Qunying, Scientist.

Another development is the use of artificial intelligence (AI) to automate rotifer counting and observe rotifer populations. Daily monitoring of rotifers under a microscope is routine to ensure health and stability of rotifer cultures. This requires the counting of the number of eggs, observation of rotifer activity and movement, and ciliate contamination. The whole process takes up to 40 minutes and can be subjective.

Xu Qunying described a new AI software which gives a quicker and objective assessment of rotifer numbers and quality. The platform captures images of the rotifer culture, identifies the rotifer and ciliates and enables sharing of information across partners on the cloud. Furthermore, it enables digitalisation of routine operations and reduces dependency on experience for assessments. "In the future, we can further automate rotifer production through development of deep learning technology platform to determine the health of the rotifer culture for continuous and reliable production to support fish hatcheries."

Discussing complexities of manual counting of rotifers



Liang Bing describing the marker assisted selection programme to visitors in conjunction with the opening of ASA (see pages 60 and 62)

First in milkfish farming in Malaysia

With its commercialisation achieved, Usaha Fadzilat next looks at creating appetites for this novel fish among Malaysian consumers.

In 2014, Lim Yong Keng, Manager at Usaha Fadzilat (M) Sdn Bhd, came across the milkfish *Chanos chanos*, which stirred his curiosity and led to his 5-year journey on adapting the farming technology and management to a commercial scale in the 200 acres (80.4 ha) farm with 31 ponds in Batu Lintang, Sungai Petani, Kedah. The milkfish is a popular fish in Indonesia, the Philippines and Taiwan where the annual production was 778,502 tonnes in Indonesia (Dahuri, 2019) and 238,940 tonnes in the Philippines (2019, psa.gov.ph). In Malaysia, the fish is relatively unknown. However, there are some recent reports of occasional farming of the fish together with the Asian seabass or vannamei shrimp.

At the launch of this new business in January, which was attended by officials from the Department of Fisheries (DOF), Malaysia and the aquaculture and seafood industry in Malaysia, Yong Keng, said, "Many have asked why did we decide on the milkfish. We believe that this fish species will benefit local consumers as it is known for its high omega-3 fatty acids and protein content. It is a herbivorous fish and easy to farm. We have worked hard to study its production technology and after 5 years, I am here to announce 2019 as our first year of commercial production. Usaha Fadzilat has successfully produced a total of 3,000 tonnes of milkfish over the past 2 years.

Top: Lim Tio Huat (left) with Datuk Seri Salahuddin Ayub, former Minister of Agriculture and Agro Based Industries, Malaysia, at the launch in January. Lim Yong Keng is second left.

Bottom: Promoting Usaha Fadzilat's deboned milkfish at an exhibition in Penang with the Director General of the Department of Fisheries, Dato' Haji Munir Mohd Nawi (right) with DOF officials as well as Ching Kok Ying (back, third left).

View of the 1.6ha milkfish ponds surrounded by the long reservoir

Harvesting milkfish after 6 months of culture.

"Of course, we are still very far from the 700 tonnes/day production in the Philippines, but we are just starting. With continuous development and perfecting our farming technology, we are confident that we can reach this target in the foreseeable future. We anticipate our milkfish production will grow. I look forward to taking our milkfish farming business to the global market in 10 years with a production of 30,000 tonnes/year." Together with Yong Keng in this new venture is Ching Kok Ying, the company General Manager. Ching, an expert in aquaculture, played a key role with his 10 years in steering the company, firstly in farming the vannamei shrimp and now in milkfish farming.

Usaha Fadzilat has achieved success as the first in Malaysia to reach this level of production, according to the press release by the DOF, Malaysia. The company has accreditation to MYGaP (Malaysia Good Aquaculture Practices) and Fish Quality Certificate (FQC), issued by DOF.

From shrimp to milkfish

Usaha Fadzilat has been in the Malaysian aquaculture scene for 32 years. It was started by Yong Keng's father, Lim Tio Huat,

Milkfish harvested from ponds range from 500-800g. The daily harvest is sold to local markets in Kedah, Penang and Selangor. Deboned ikan susu, as the milkfish is called in Malaysia has a 20% price premium.

who began by farming the monodon shrimp and subsequently moved to farm the vannamei shrimp. The elder Lim, now the chairman, has passed the running of the farm to his two sons, Yong Keng and Shen Yong, some 1.5 years ago. Today, this younger generation plays a larger role in managing the farm. The farm has 50 workers for the farming segment.

"We decided to switch to the milkfish in 2014 and each crop cycle is 6 months. We did not need to renovate the shrimp ponds extensively. In this farm, our 4 acre-ponds (1.6 ha) are enclosed by two long reservoir ponds of 3m deep which follow the loop of the river. We draw the water upstream from the river at one end and release water downstream into the same river at another end. Water is filtered with an 80-mesh net and we exchange almost all of the water daily. Water salinity is around 25-28ppt, ideal for the milkfish," said Yong Keng.

"One of our early challenges was to formulate a specific feed for the milkfish. We worked with Cargill Malaysia to develop a 26-28% crude protein (CP) feed. Next is to have a lower protein feed of 24% CP. Our feed demand is 300 tonnes/month. Currently the cost of production is MYR6/kg (USD1.4/kg) at a feed conversion ratio of 1.6. The average survival rate is 60%, which is acceptable. Margins are good today as we are the only producer." The good news is that Yong Keng has estimated a survival rate of 80% for this year's first crop.

Marketing milkfish in Malaysia

As the milkfish is a new product in Malaysia's seafood scene, marketing the fish is a major challenge for the brothers. Yong Keng explained, "Our competitors are the farmed seabass and snappers, which local consumers are familiar with. Now we harvest daily and sell the fish as chilled or fresh fish in markets in Selangor, Penang and Kedah. We named the fish as 'ikan susu' in Bahasa Malaysia or 奶宝鱼 in Mandarin. Acceptance of live fish is still challenging as restaurants prefer seabass etc. for steamed fish dishes. The ex-farm price is MYR10/kg (USD2.4/kg) whereas at the retail level, the fish is sold at MYR18-19/kg (USD4.25-4.5/kg). We also export the fish through a broker, currently to China and Australia."

He added, "We want Malaysians to appreciate the benefits of this 'vegetarian' fish. We have been attending exhibitions to promote the fish. At this launch, we have prepared several dishes to introduce western, oriental, fried and steamed milkfish recipes. We have recently started an e-commerce business (fishco.com.my) as another channel to market the milkfish."

The milkfish is well known as an extremely bony fish. The team then sourced experts from Taiwan and the Philippines on techniques to remove all 222 bones from the fish. The company's processing plant was started in September 2019 and daily 40 workers manually remove the fish bones. "We strongly believe that our success in deboning milkfish has increased its acceptability and popularity in the Malaysian market. Deboned milkfish, vacuum packed and frozen, has a 20% price premium. Next we can look further to value added products such as smoked milkfish and fish ball."

Similar to the tilapia, Yong Keng is looking at the farm fish complex, apart from the fillet for food, the rest of the fish for various co-products. "We know that we can use the bones as fertiliser, scales for face masks and now we are working with chefs to develop the skin into snacks."

Lim Tio Huat (centre) and Lim Yong Keng (third right) with the Cargill Asean team.

Expansion: hatchery and contract farming

Currently, the farm buys 1-inch (2.54 cm) fry for stocking from Taiwan and the Philippines. However, there are plans to have its own hatchery. It already has around 700 broodstock of 3-10kg awaiting the set-up of a hatchery. "We are hoping to share our farming knowledge with farmers who are interested in starting a milkfish farming business under our guidance and using our farming technology and management practices," added Yong Keng.

2020 Global Feed Survey

The 2020 Alltech Global Feed Survey estimates the global animal feed production has declined by 1.07% to 1.126 billion tonnes in 2019, with the top nine countries producing 58% of this production. This is the first drop in world feed production since the survey began 9 years ago.

The 2020 Alltech Global Feed Survey estimates that global animal feed tonnage decreased by 1.07% to 1.126 billion tonnes of feed produced in 2019, due largely to African swine fever (ASF) and the decline of pig feed demand in the Asia-Pacific region. The top nine feed-producing countries are the U.S., China, Brazil, Russia, India, Mexico, Spain, Japan and Germany. Together, these countries produce 58% of the world's feed production where 57% of the world's feed mills are located, and they can be viewed as an indicator of overall trends in agriculture.

Annually, this feed survey assesses compound feed production and prices through information collected in the last quarter of 2019 by Alltech's global sales team in partnership with local feed associations. The data serve as a resource for policymakers, decision-makers and industry stakeholders. Dr Mark Lyons, president and CEO of Alltech, shared the survey results via public livestream from the company's global headquarters in Nicholasville, Kentucky.

"2019 presented extreme challenges to the feed industry, with one of the most significant being AFS. The regional and global implications are reflected by the Alltech Global Feed Survey and the decline in global feed production," said Lyons. "While pig feed production is down in affected countries, we are noting increased production both in other species as producers work to supplement the protein demand, and in non-affected countries as exports ramp up. The damage caused by ASF will have long-term implications, and we expect that the top protein sources will continue to shift as our industry adapts to the shortage."

Lyons also added the reason Alltech conducts this annual survey. "Every year we look at the results and ask our colleagues in September to October to look at the trends. We do this because it is an annual barometer on the livestock feed sector, an indicator of economic growth. It also serves as a database for governments, industry groups and associations." The data was collected in two parts; quantitative numerical data and qualitative with short answer responses. In the latter, for aquaculture, the question was on trends.

The global data, collected from 145 countries and nearly 30,000 feed mills, indicated feed production by species as: broilers 28%; pigs 24%; layers 14%; dairy 12%; beef 10%; other species 6%; aquaculture 4%; and pets 2%. Predominant growth came from the layer, broiler, aqua and pet feed sectors.

Regional results from this 2020 feed survey showed the following. In North America, the US is the largest feed-producing country globally with an estimated 214 million tonnes, an increase of 1.6% from the 2018 figures. As a region, Latin America saw 2.2% growth to 167.9 million tonnes led by Brazil which was also third overall globally. The primary species for feed production was broilers at 32.1 million tonnes and pig at 17.0 million tonnes. Brazil, Mexico and Argentina continued to produce most of the feeds in Latin America comprising 76% of regional feed production.

However, European feed production remained relatively stagnant with a slight increase of 0.2% over that in 2018. The top three feed-producing countries in Europe were: Russia (40.5 million tonnes), Spain (34.8 million tonnes) and Germany (25.0 million tonnes), with pig feed production leading the way in all three countries. There was a strong growth in aquafeed production at 7% and feed for layers at 3%.

The Asia-Pacific region saw feed production decreased by 5.5% in 2019, primarily due to AFS and large declines in pig feed production. China's feed production declined by almost 20 million tonnes overall to 167.9 million tonnes. China fell from its top position to be second globally, behind the US. India and Japan remained in the top nine feed-producing countries, with production similar to 2018 at 39.0 million tonnes and 25.3 million tonnes, respectively, while Vietnam's production declined by 7%.

Africa continued its strong growth with a 7.5% increase in overall feed production, with all the primary species seeing growth. The top five feed-producing countries in the region accounted for 75% of Africa's feed production, and they were South Africa, Egypt, Nigeria, Morocco and Algeria. The region's primary species included broiler, layer and dairy; combined, they accounted for nearly half of the feed production estimates in the region.

With AFS, the Asia-Pacific region was significantly impacted with pig feed production down 26% while protein alternatives were up, such as layer (7%) and broiler (6%). The most affected countries were China, Cambodia, Vietnam, Thailand, the Philippines and Mongolia. Europe was also affected, primarily Estonia, Latvia, Lithuania, Romania, Ukraine and Bulgaria with higher pig prices (20%).

Global aquafeed production

Overall, the 2019 global aquaculture feed production showed growth of 4% over that in 2018 to 41 million tonnes. Asia-Pacific production grew the most with an additional 1.5 million tonnes reaching 30 million tonnes. Latin America produced 4.2 million tonnes led by Brazil with 1.260 million tonnes, Ecuador with 650,000 tonnes and Mexico with 419,000 tonnes. Europe was next with 3.8 million tonnes where Norway was the leading aquafeed producer at 1.624 million tonnes, followed by Turkey (408,000 tonnes) and Greece (220,000 tonnes). Europe's decrease was in large part due to decreased feed production in Russia, which was primarily due to an increase in imports.

The US and Canada produced 1.710 million tonnes of aquafeeds. In the Middle East and Africa, the leading producer was Egypt at 800,000 tonnes and the largest producer in the Middle East was

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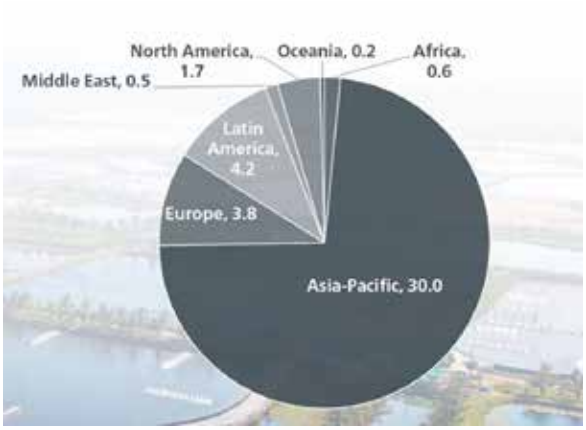

Figure 1. Global aquafeed production in tonnes presented at the 2020 Alltech Global Feed Survey in January 2020.

Iran with 280,000 tonnes and next was Saudi Arabia at 80,000 tonnes. Smaller producers at 10,000 tonnes or more included Oman (30,000 tonnes), with Zambia, Zimbabwe and United Arab Emirates, each producing 10,000 tonnes annually.

Asia Pacific aquafeed production

This 2020 global survey showed that in the Asia Pacific region, the primary contributors were China, Vietnam, India and Indonesia (Figure 2). China produced 16.527 million tonnes, accounting for 55% of the production in the Asia Pacific and 41% of global aquafeed production. Comparing this data from that of the 2019 survey, both China and Vietnam increased production by 5%; India increased production by 8.57% and Indonesia by 4.98%. Together, these four countries contributed an additional 1.25 million tonnes of feed to the region. The survey continued to present aquafeed production from smaller emerging producers, including in Asia Pacific: Laos, Cambodia, Pakistan and Sri Lanka (see. Table 1).

During the live presentation, Lyons was joined by a panel of industry experts, including Jack Bobo, CEO, Futurity, USA; Matthew Smith, vice president, Alltech, U.K.; Bianca Martins, general manager, Alltech, Mexico; and Brian Lawless, North America species manager, Alltech, USA. The group discussed the trends behind the data and the implications for the global market. Topics ranged from consumer demands to the adoption of new technology. They also discussed what changes the industry may expect within the next year and beyond.

Insights from the 2020 Alltech Global Feed Survey, including a recording of the panel discussion, an interactive map and presentation slides, are available at alltechfeedsurvey.com. Scan the QR code on above right for the interactive map.

Country	Tonnes in 2019
China	16,527,000
Vietnam	4,070,000
India	2,280,000
Indonesia	1,877,000
Philippines	1,400,000
Thailand	1,010,000
Bangladesh	1,000,000
Taiwan	442,000
Japan	550,000
Myanmar	370,000
Korea	200,000
Malaysia	150,000
Sri Lanka	19,000
Pakistan	15,000
Cambodia	10,000
Laos	6,000

Table 1. 2019 Aquafeed production in Asia Pacific (source: 2020 Alltech Global Feed Survey).

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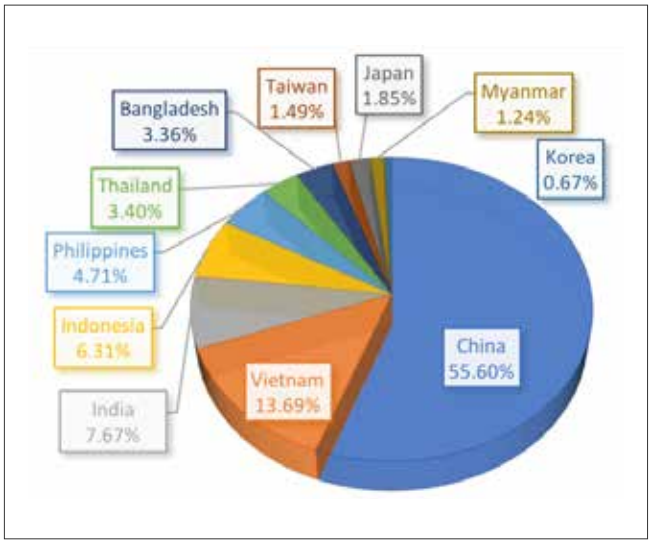

Figure 2. The top aquafeed producing countries in Asia Pacific and percentage contribution to the total 2019 production of 30 million tonnes (source: 2020 Alltech Global Feed Survey).

L-selenomethionine: A powerful antioxidant for use in commercial fish culture

Trials with tilapia in Thailand showed increased performance and high protection against pathogenic pressure

by Brecht Bruneel

In intensive animal production, high daily weight gain and high feed efficiency are essential. However, high performance is associated with increased levels of stress. Stress such as from high stocking density, pathogenic pressure and temperature is associated with enhanced levels of reactive oxygen species (ROS) and linked to suboptimal antioxidant status. Selenium (Se), in this respect, is a very important essential trace element as it is a vital component of selenoenzymes (e.g. glutathione peroxidase, GPx) which play a role in reducing ROS and in maintaining a healthy antioxidant status. A disruption of this steady state causes tissue damage due to interaction of ROS with lipids, proteins and DNA. These negative interactions reduce their metabolic activity.

In order to maintain this steady state a continuous as well as optimal selenium supply is essential. However, this can be difficult to achieve when uptake from the diet is impaired when stress is present. At that moment selenium is in high demand, to produce selenoenzymes and combat ROS. Selenium storage inside the animal, in that respect, would be beneficial. This article provides an overview of the scientific literature on the beneficial effects seen with the addition of L-selenomethionine to the diet with focus on salmon, trout and tilapia. Results from a recent trial on tilapia conducted in Thailand are discussed.

Maintaining an optimal selenium steady state: A nutritional solution

Selenium can be added to the diet in either inorganic or organic forms (Figure 1). The advantage of using organic selenium (L-selenomethionine, L-SeMet) over inorganic sources (e.g. sodium selenite or selenate) is its ability to be incorporated directly, without conversion, into general body proteins as a methionine source. L-selenomethionine is the only selenium compound that has this ability. The incorporated selenium, in the form of L-selenomethionine, acts as a storage of selenium in the animal. This stored selenium ensures optimal supply, even during stressful periods.

If necessary, the stored selenium gets metabolised to selenide (H_2Se) then to *de novo selenocysteine* (SeCys). This molecule will be incorporated, as the active site, in selenoproteins. Other selenium compounds, such as SeCys and sodium selenite, are not storable but will be metabolised to de novo SeCys. These compounds will be quickly excreted when intake is in excess. L-selenomethionine will only be metabolised to selenide when there is a need. This form is therefore less prone to excretion and toxicity reactions (Rayman, 2004).

Aquatic protein challenge: A case for L-selenomethionine

Traditionally, fishmeal is the preferred protein source in aquatic feeds. Due to limited availability, pressure on wild fish stocks and variable prices, there is an interest in alternative, sustainable protein sources. Plant meals, for example, are suitable alternatives in the growing global aquaculture industry. However, replacing marine ingredients in fish feed with plant sources changes the nutrient composition of the feed. Selenium concentration of fillets is reported to be highly impacted by high levels of substitution, reducing the added value of fish consumption (Lundebye et al. 2017; Betancor et al. 2016).

Although selenium levels are decreasing within the fish, the demand for selenium to protect against (oxidative) stress remains. Stressors (e.g. environmental, metabolic) are an important issue for the productivity and profitability of fish farms. These stressors may cause increased oxidative damage to lipids, proteins and DNA and increased mineral mobilisation from tissues and their subsequent excretion. High stress may therefore lead to increased mineral requirement. L-selenomethionine is established to be a highly available selenium source leading to higher selenium deposition compared to inorganic selenium sources (Figure 2). It can therefore counteract selenium depletion caused by plant-based diets.

Control stress and win!

Dietary selenomethionine supplementation is known to offer a way to reduce performance loss under stress, such as crowding conditions (Küçükbay et al. 2008). A recent study, performed at the Mahasarakham University, Thailand, showed increased performance and high protection against pathogenic

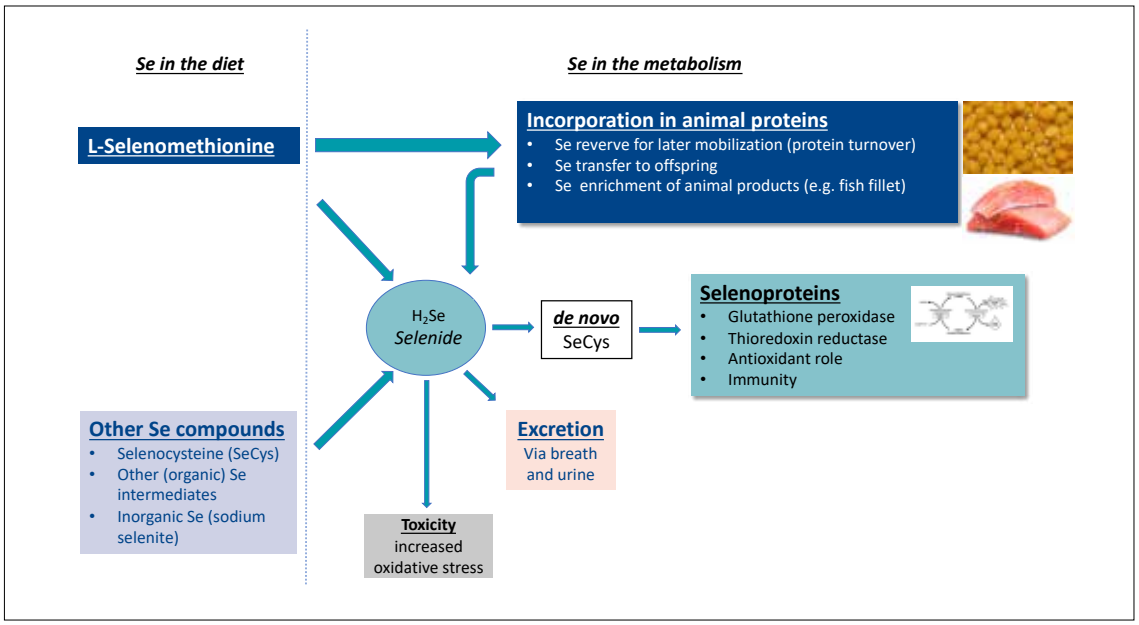

Figure 1. The metabolism of L-selenomethionine and other selenium compounds (adapted from Rayman, 2004; and Combs, 2001).

pressure. A total of 735 Nile tilapia (initial weight 13.52±0.5g) were fed one of seven experimental diets (in triplicate) in fiberglass tanks for 8 weeks. Organic Se (L-selenomethionine, SeMet; Excential Selenium 4000, Orffa Additives BV) and inorganic Se (sodium selenite, Na_2SeO_3) were each added to the basal diet at 1, 3, and 5mg Se/kg. The basal diet (28% crude protein), without Se supplementation, was used as a control.

The final Se concentration of the basal diet was 0.68mg Se/kg. Organic and inorganic Se supplemented diets contained 1.78, 3.53 and 4.90mg Se/kg and 1.75, 3.49 and 5.30mg Se/kg, respectively. Fish were fed at 5.0% of their body weight twice a day. Parameters were assessed at the end of the rearing period. After 8 weeks, 20 fish from each treatment were challenged with intraperitoneal injection of the virulent *Streptococcus agalactiae* serotype III at $1x10^7$ CFU/mL. The cumulative mortality was observed for 21 days and the relative percent survival (RPS) was calculated.

Table 1 shows that weight gain (WG) of fish fed SeMet at 1mg Se/kg was significantly higher than that of fish fed basal diet ($p<0.05$). Lymphocytes were significantly ($p<0.05$) higher in fish fed SeMet (1mg Se/kg) compared to fish fed basal diet. Alanine transaminase (ALT), aspartate transaminase (AST), creatinine, blood urea nitrogen (BUN), albumin, globulin and total protein were not significantly influenced by dietary Se supplementation.

Increasing dietary Se level, particularly in the form of SeMet, led to a decrease in serum cholesterol concentrations. Interestingly, the innate immune response (e.g. lysozyme, catalase, myeloperoxidase, superoxide dismutase and glutathione peroxidase) activity was significantly ($p<0.05$) increased with Se supplementation compared to the basal diet group, especially for fish fed SeMet (1 and 3mg/Se kg). Malondialdehyde (MDA) in fish serum, a clear biomarker for oxidative stress, decreased numerically for all supplementation levels. Fish fed SeMet (1mg Se/kg) showed the highest RPS after the challenge with *S. agalactiae*.

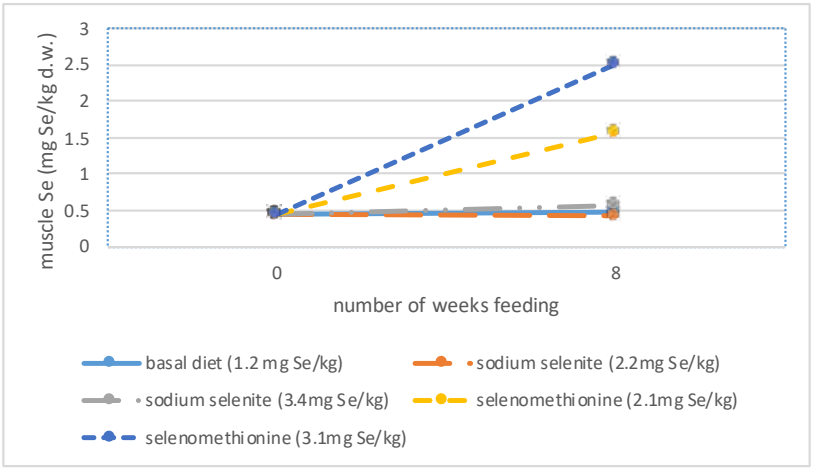

Figure 2. Selenium concentrations in muscle (mg Se/kg dry weight) of Atlantic salmon fed a fishmeal-based diet supplemented with sodium selenite or selenomethionine at levels of 1 and 2 mg Se/kg feed, respectively, for 8 weeks (Lorentzen et al. 1993).

Conclusions

L-selenomethionine (Excential Selenium 4000) was tested and validated by independent researchers around the world in peer-reviewed publications (e.g. Berntssen et al. 2018; Silva et al. 2019) and proven to be effective in increasing the selenium and antioxidant status of fish, even under challenging conditions. This will result in improved performance and immune function. Very high levels of L-selenomethionine (5mg Se/kg feed) do not appear to have negative effects on performance nor on immune parameters.

L-selenomethionine therefore has a good application in fish diets when fish are kept under stressful conditions or in any diets where fishmeal is replaced by plant meals. L-selenomethionine helps to maintain selenium concentration in fish fillets and therefore contributes to the positive healthy image of fish in human nutrition.

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	Basal diet (total mg Se/kg)	L-SeMet treatments (total Se/kg feed)			Sodium selenite treatments (total Se/kg feed)			Pooled SEM
Parameter	**0.68**	**1.78**	**3.53**	**4.90**	**1.75**	**3.49**	**5.00**	
Weight Gain (g)	40.36 b	53.62 a	47.98 ab	39.34 b	40.51 ab	43.07 ab	39.02 b	2.01
Feed conversion ratio (FCR)	1.77 ab	1.42 a	1.54 ab	1.79 ab	1.75 ab	1.67 ab	1.82 b	0.1
WBC (cells/mm^3)	2,275.5 ab	2,849.0 a	1,859.8 b	2,109.0 ab	2,109.0 ab	2,026.3 ab	1,887.0 b	108.89
Lymphocytes (%)	39.3 bc	49.7 a	42.6 abc	40.5 abc	48.0 ab	41.5 abc	36.0 c	3.6
ALT (U/L)	22.0 a	17.6 ab	19.67 ab	14.0 ab	16.33 ab	13.4 b	15.0 ab	2.93
AST (U/L)	53.33 a	51.4 ab	50.67 ab	61.67 a	48.6 b	67.5 a	66.2 a	10.3
BUN (mg/dL)	2.0	1.0	1.3	1.3	1.7	1.0	1.3	0.27
Albumin (g/dL)	1.03 ab	1.17 ab	0.93 b	1.00 ab	1.03 ab	1.20 a	1.10 ab	0.07
Globulin (g/dL)	2.3	2.3	2.1	2.3	2.3	2.3	2.2	0.07
Total protein (g/dL)	3.33 ab	3.50 a	3.03 b	3.27 ab	3.33 ab	3.47 a	3.27 ab	0.15
Cholesterol (mg/dL)	175.67 a	153.67 ab	125.67 b	159.67 a	157.00 a	161.0 a	152.33 ab	9.31
Lysozyme activity (U/mL)	12.50 d	30.25 a	23.75 b	17.67 c	12.80 d	25.00 b	17.00 c	3.82
Catalase activity (U/mL)	6.67 d	20.00 a	13.13 bc	6.67 d	11.25 c	15.63 b	3.25 d	4.42
Myeloperoxidase (OD at 450)	0.70 d	1.16 a	1.13 ab	1.05 abc	0.83 cd	0.71 d	0.71 d	0.10
Superoxide dismutase (U/mL)	39.19 c	47.81 a	43.65 bc	42.75 bc	42.43 bc	45.66 ab	42.75 bc	2.97
Glutathione peroxidase (mU/mL)	15.13 b	38.91 a	27.23 ab	20.32 b	30.4 a	36.2 a	16.75 b	3.40
MDA (mmol/mg protein)	130.12 a	116.39 a	114.24 a	111.96 a	98.29 a	102.76 a	115.71 a	7.30
RPS (%)	-	84.62	53.85	46.15	30.77	53.85	23.08	-
Glutathione peroxidase (mU/mL)	15.13 b	38.91 a	27.23 ab	20.32 b	30.4 a	36.2 a	16.75 b	3.40
MDA (mmol/mg protein)	130.12 a	116.39 a	114.24 a	111.96 a	98.29 a	102.76 a	115.71 a	7.30
RPS (%)	-	84.62	53.85	46.15	30.77	53.85	23.08	-

Table 1. Growth performance, haematological values and immune parameters of fish fed experimental diets. Note: Values show mean, pooled SEM, n = 90; S= L-SeMet (Excential Selenium 4000) and sodium selenite (Na_2SeO_3), L=level of selenium supplementation. Values in the same row with different letters differ significantly ($p < 0.05$). White blood cells (WBC), alanine transaminase (ALT), aspartate transaminase (AST), blood urea nitrogen (BUN), malondialdehyde (MDA), relative percent survival (RPS).

References

Rayman, M.P. 2004. The use of high-selenium yeast to raise selenium status: how does it measure up? British Journal of Nutrition. 92, 557-573

Combs, G.F. 2001. Selenium in global food systems. British Journal of Nutrition. 85, 517-547

Lundebye, A., Lock, E., Rasinger, J.D., Nøstbakken, O.J., Hannisdal, R., Karlsbakk, E., Wennevik, V., Madhun, A.S., Madsen, L., Graff, I.E., Ørnsrud, R. 2017. Lower levels of Persistent Organic Pollutants, metals and the marine omega 3-fatty acid DHA in farmed compared to wild Atlantic salmon (*Salmo salar*). Environmental Research. 155, 49-59

Betancor, M.B., Dam, T.M.C., Walton, J., Morken, T., Campbell, P.J., Tocher, D.R. 2016. Modulation of selenium tissue distribution and selenoprotein expression in Atlantic salmon (*Salmo salar* L.) fed diets with graded levels of plant ingredients. British Journal of Nutrition. 115, 1325-1338

Lorentzen, M., Maage, A., Julshamn, K. 1994. Effects of dietary selenite or selenomethionine on tissue selenium levels of Atlantic salmon (*Salmo salar*). Aquaculture. 121, 359-367

Küçükbay, F.Z., Yazlak, H., Karaca, I., Sahin, N., Tuzcu, M., Cakmak, M.N., Sahin, K. 2009. The effects of dietary organic or inorganic selenium in rainbow trout (*Oncorhynchus mykiss*) under crowding conditions. Aquaculture Nutrition. 15, 569-576

Berntssen, M.H.G., Betancor, M., Caballero, M.J., Hillestad, M., Rasinger, J., Hamre, K., Sele, V., Amlund, H., Ørnsrud, R. 2018. Safe limits of selenomethionine and selenite supplementation to plant-based Atlantic salmon feeds. Aquaculture. 495, 617-630

Silva, M.S., Kröckel, S., Prahbu, P.A.J., Koppe, W., Ørnsrud, R., Waagbø, R., Araujo, P., Amlund, H. 2019. Apparent availability of zinc, selenium and manganese as inorganic metal salts or organic forms in plant-based diets for Atlantic salmon (*Salmo salar*). Aquaculture. 503, 562-570

Better growth performance with a multi-enzyme supplementation

Trials show that a multi-enzyme supplementation allows complete fishmeal replacement in diets for red tilapia and walking catfish

By Rutchanee Chotikachinda, Viviane Verlhac Trichet and Fidelis Fru

Today, in aquaculture nutrition, there is added focus on non-fishmeal protein for feeds and in finding alternative protein sources. Fishmeal can be replaced by plant and land animal protein sources which will reduce aquaculture's impact on overfishing and prevent the creation of an imbalance in the marine ecosystem. The formulation of feeds with alternative protein ingredients is more environmentally balanced and more economical with lower fishmeal levels; it also reduces supply chain risks from external factors like climate change and drives progress in aquafeed development.

Feed enzymes

However, the use of a wider range of feed raw materials makes the diets more complex and increases the variability in its nutritive value and digestibility. Feed enzymes are one effective way of improving feed utilisation and enhancing bioavailability of these alternative feed ingredients. To increase the nutrient utilisation from many feedstuffs, the supplementation of complex diets with multi-enzyme mixtures has additive effects (Dai et al., 2018) is effective in improving dietary nutrient digestibility (Zheng et al., 2019), leading to better feed performance.

Major ingredients in aquafeed are fishmeal, processed animal proteins and agriculture ingredients such as vegetable proteins (e.g. soy), cereals and grain products. For various reasons, fishmeal and animal protein are gradually being replaced by plant proteins in aquafeeds. This leads to changes in protein and amino acid digestibility, increased content of antinutritional factors and imbalances in nutrient composition. The potential of multi-enzyme application to increase the availability of nutrients in high plant-based diets is expected to be high when ingredients with significant concentrations of indigestible substrates and poor nutrient availability are used in aquafeeds.

Primarily phytase has been used to breakdown phytic acid, thus increasing available phosphorus, and freeing phytate-bound trace minerals and protein in plant-based feeds. Research has shown that the digestibility of protein and energy from alternative ingredients may be limited in some species and phytase cannot eliminate all the problems associated with plant ingredients. Consequently, utilising non-starch polysaccharides (NSP) degrading enzymes to reduce caged effects of insoluble NSP and the viscosity of soluble NSP has been found to be beneficial for fish. Indeed, these enzymes improve the practicality and economy of using plant proteins as alternative proteins in feed formulations.

The application of multi-enzymes to improve nutrient digestibility and utilisation, places importance on selecting enzymes with specific activity matching the substrates found in ingredients. In the present study, the advanced enzyme combinations (RONOZYME®) shown in Table 1 were designed for use with complex aquafeed formulations for numerous species, but the specific response to enzymes by tilapia (*Oreochromis niloticus*) and walking catfish (*Clarias macrocephalus* x *Clarias gariepinus*) was investigated here.

Advanced enzyme combination

DSM has the widely used phytase (RONOZYME® HiPhos) in aquafeed which functions by hydrolysing dietary phytic acid into absorbable forms of phosphorus (P), while simultaneously reducing the anti-nutritional effects, resulting in more efficient use of amino acids, trace minerals and energy. A protease (Ronozyme® ProAct), effectively degrades proteins to amino acids through its non-specific hydrolysis of peptides, enhancing protein digestibility across various marine and terrestrial protein sources. Protease also exposes phytic acid trapped in protein globules (e.g. soy protein) making it susceptible to the action of a phytase. Use of several plant cell wall degrading enzymes (Ronozyme® WX, Ronozyme® MultiGrain and Ronozyme®VP) reduces the antinutritional effects of NSP. Each targets a specific NSP substrate. In addition, the xylanase enzyme debranches arabinoxylan, decreasing NSP viscosity, while exposing nutrients trapped in plant cells, to other digestive enzymes. A combination of fibre degrading enzymes help to break down cell walls by solubilising xyloglucan, revealing an inner pectin layer which can be hydrolysed by a pectin hydrolysing enzyme (Ronozyme®VP).

> "The enzyme supplementation contributes to the replacement of 3% fishmeal in the red tilapia diet and 5% fishmeal in the walking catfish diets, and compensates for the reduction inorganic phosphate uses and digestible protein in feed formulation."

Table 1. Advanced combinations of feed enzymes used in tilapia and walking catfish growth performance trials.

Enzymes	ESol1 (unit/kg feed)	ESol2 (unit/kg feed)	ESol3 (unit/kg feed)
HiPhos (GT)	1,000 FYT	2,000 FYT	1,500 FYT
WX 2000 (CT)	100 FXU	250 FXU	50 FXU
ProAct (CT)	15,000 PROT	30,000 PROT	3,750 PROT
VP (CT)*			+
MultiGrain (CT)**			+

* Multicomponent carbohydrase for non-cereal product
**Multicomponent carbohydrase for cereal and their by-product
FYT = phytase activity unit (One unit of phytase activity is the amount of enzyme which release 1 micromole inorganic phosphate per minute from a 0.0051 M Na-phytate solution pH 5.5 and 37˚C)
FXU = xylanase activity unit (One unit of xylanase activity is the amount of enzyme that releases 1 micromole of reducing moieties from 1.5% arabinoxylan substrate solution per minute at pH 5.0 and 40˚C)
PROT = protease activity unit (one protease unit is the amount of enzyme that release 1 micromole of p-nitroaniline from 1 millimolar substrate per minute at pH 9.0 and 37°C)

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DSM

In this research, we focused on using multi-enzymes with fishmeal replacement in the diet formulation to investigate the effects of the enzyme combination on growth performance of two species, red tilapia and walking catfish. The red tilapia trial was carried out at CRNA, DSM Nutrition Products in France while the walking catfish trial was conducted at Thaksin University Patthalung Campus in Thailand by Dr Suphada Kiriratnikom.

Experimental design for red tilapia

A 2 by 3 factorial design was performed to evaluate the influence of fishmeal inclusion (factor 1) and enzyme supplementation (factor 2) on growth performance and feed utilisation in red tilapia. The fishmeal diet (FMD) and non-fishmeal diet (NFMD) were formulated to meet the nutritional requirement of tilapia with inorganic P supplementation and the addition of enzyme combinations ESol1 and ESol2, containing phytase, xylanase and protease as described in Table 1. The formulation of basal diets (FMD and NFMD) and the analysed chemical composition of the experimental diets are presented in Table 2 and 3, respectively. Twenty-five fish with an average initial body weight of 56.9±0.1 g were stocked in 18 tanks (three replicate tanks per treatment), with waters kept at 28˚C and supplied from a recirculating water system. The fish were fed three times per day at the fixed rate of 4-5% of their body weight for 8 weeks to final measurement.

The addition of multi-enzyme combinations ESol1 and ESol2 to the FMD and NFMD diets resulted in increased weight gain compared with fish that were fed

Ingredients	FMD	NFMD
Fishmeal	3	0
Poultry meal	12	10
Wheat, middling	15	15
Corn grain	20	20
Soybean meal	22	30
Wheat flour	5	5
Corn gluten meal	5	5
Rice bran	14.57	11.53
Fish oil	1	1
Soybean oil	1	1
Choline chloride	0.6	0.6
Vitamin & Mineral	0.05	0.05
L-Lysine	0.2	0.2
DL-Met	0.18	0.22
DCP	0.4	0.4
Total	100	100

Table 2. Diet formulations for red tilapia.

Experiment diets	FMD	FMD+ESol1	FMD+ESol2	NFMD	NFMD+ESol1	NFMD+ESol2
Crude protein(%)	32.20	32.80	31.90	32.50	32.10	32.50
Crude lipid (%)	7.70	7.50	7.60	6.70	6.70	6.90
Crude ash (%)	5.82	5.96	5.97	5.47	5.44	5.53
Dry matter (%)	91.70	92.30	92.00	91.90	92.10	92.20
Energy (MJ/kg)	18.86	18.98	19.00	19.05	18.70	19.03
Total P (%)	0.97	0.99	0.99	0.89	0.88	0.87

Table 3. Proximate composition of experimental diets for red tilapia.

Dietary treatments	Survival rate	FCR	SGR (%)
FMD	100.00 ± 0.00	1.29 ± 0.03	2.30 ± 0.05
FMD+ESol1	98.67 ± 2.31	1.27 ± 0.04	2.33 ± 0.06
FMD+ESol2	100.00 ± 0.00	1.26 ± 0.02	2.34 ± 0.04
NFMD	100.00 ± 0.00	1.43 ± 0.14	2.11 ± 0.17
NFMD-ESol1	100.00 ± 0.00	1.24 ± 0.13	2.40 ± 0.23
NFMD-ESol2	98.67 ± 2.31	1.25 ± 0.02	2.35 ± 0.03

Table 4. Survival rate, FCR and SGR of red tilapia fed different diets for 8 weeks.

Parameters	P value		
	Fishmeal inclusions	Enzyme solutions	FM*ENZ interaction
Final body weight	0.056	0.001	0.719
Survival rate	1.000	0.619	0.262
FCR	0.014	0.000	0.157
SGR	0.046	0.001	0.705

Table 5. *P* value for factorial analysis.

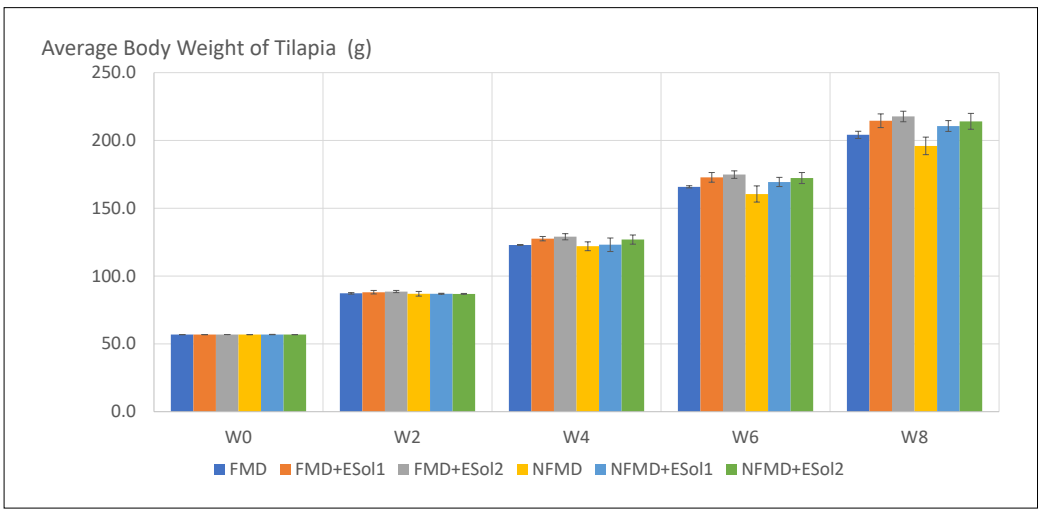

Figure 1. Body weight (BW) of red tilapia fed diets with different inclusions of fishmeal and levels of enzyme for 8 weeks (p >0.05 after week 6).

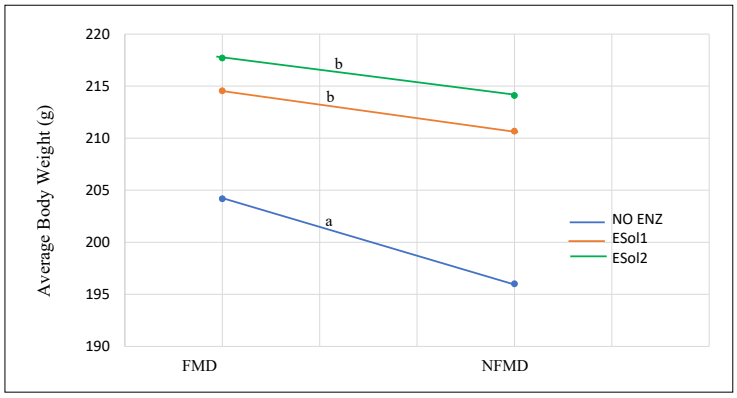

Figure 2. Average body weight comparing two factors, fishmeal inclusions (p> 0.05) and enzyme solutions (p<0.05)

non-enzyme supplemented basal diets ($p < 0.05$). The benefit of multi-enzyme supplementation was observed after the 6-week feeding period, with a significant increase in growth performance of red tilapia (Figure 1). Figure 2 shows the beneficial effect of ESol1 and ESol2 compared between FMD and NFMD.

The factorial analysis indicated a significant effect of enzyme combinations for the NFMD ($p<0.05$), but no significant effect of enzyme inclusion on the FMD on growth performance was observed ($p>0.05$). There is no evidence of the interaction of fishmeal inclusions and enzyme solutions ($p >0.05$) among the dietary treatments. After 8-weeks, there were clear differences in specific growth rate (SGR) of fish fed diets supplemented with both enzyme solutions compared with those of fish fed both basal diets (Table 4 and 5). ESol1 and ESol2 showed significantly improved feed conversion ratios (FCR) when both were supplemented in FMD and NFMD ($p<0.05$).

In the tilapia study, the formulation of NFMD was done by reducing fishmeal by 3%, and poultry meal by 2% in the FMD, while soybean meal was increased to maintain protein levels. However, the multi-enzyme supplementations were likely to be equivalent in improving performance metrics in both diets. The addition of ESol1 and ESol2 from day 1 to day 56 resulted in increased weight gains of 5.1-7.5% and 6.6-9.2% respectively. Although there was a significant difference in FCRs between FMD and NFMD, supplementation with multi-enzyme solutions led to significant differences, indicating that the enzymes did not only compensate for the replacement of 5% animal protein with vegetable protein but also the variability in performance.

Ingredients	FMD	NFMD1	NFMD2
Fishmeal	5	0	0
Soybean meal	20	25	23.38
Poultry meal	20	20	18.70
Corn gluten meal	5	5	5
Corn grain	15	15	15
Cassava	15	15	15
Wheat flour	5	5	5
Wheat bran	10.38	8.93	11.84
Fish hydrolysate	1	3	3
Fish oil	0.80	0.80	0.80
Palm oil	0.80	0.80	0.80
MCP	0.55	-	-
Salt	0.50	0.50	0.50
Lysine	0.23	0.23	0.23
Methionine	0.21	0.21	0.22
OVN-FISH	0.15	0.15	0.15
Mineral premix	0.15	0.15	0.15
Mold inhibitor	0.10	0.10	0.10
Choline chloride	0.10	0.10	0.10
Carophyll® yellow	0.03	0.03	0.03
Total	100	100	100

Table 6. Formulations of experimental diets for walking catfish.

Experimental design for walking catfish

The walking catfish trial was conducted using a completely randomised design. The ESol1, which comprised the combination of three enzymes, and ESol3, the combination of five enzymes (Table 1) were incorporated into the experimental diets. The diet formulations are presented in Table 6; the dietary treatments consisted of diet 1 (FMD), diet 2 (NFMD 1), diet 3 (NFMD 2), diet 4 (NFMD 2 + ESol1) and diet 5 (NFMD1+ESol3). Each of the five experimental diets were randomly assigned to four replicates of fish and all groups were fed to satiation for 16 weeks. Growth and survival rate were monitored in each treatment group during the experimental period.

Experiment diets	FMD	NFMD1	NFMD2	NFMD2+ESol1	NFMD1+ESol3
Crude protein(%)	30.23	30.10	30.19	30.19	30.19
Crude lipid (%)	12.02	10.99	10.02	11.25	11.25
Crude ash (%)	9.52	8.23	8.56	7.46	9.51
Dry matter (%)	93.02	93.08	93.12	93.02	93.03
Energy (MJ/kg)	18.53	18.52	18.48	18.49	18.49
Total P (%)	1.78	1.14	1.22	1.34	1.31

Table 7. Proximate composition of experimental diets for walking catfish

Dietary treatments	Survival rate (%)	FCR	SGR (%)
Diet 1 (FMD)	90.40 ± 4.56	1.55 ± 0.12	2.94 ± 0.05 ^{ab}
Diet 2 (NFMD1)	92.00 ± 6.32	1.64 ± 0.18	2.86 ± 0.08 ^{a}
Diet 3 (NFMD2)	92.80 ± 3.35	1.64 ± 0.13	2.86 ± 0.04 ^{a}
Diet 4 (NFMD2+ESol1)	89.60 ± 3.58	1.57 ± 0.08	2.97 ± 0.06 ^{b}
Diet 5 (NFMD1+ESol3)	90.40 ± 8.76	1.52 ± 0.20	2.94 ± 0.06 ^{ab}

Value are means ± SD of four replicates. Means with different superscript letters in the SGR column represent significant differences (p<0.05)

Table 8. Survival rate, FCR and SGR of walking catfish fed different diets for 16 weeks

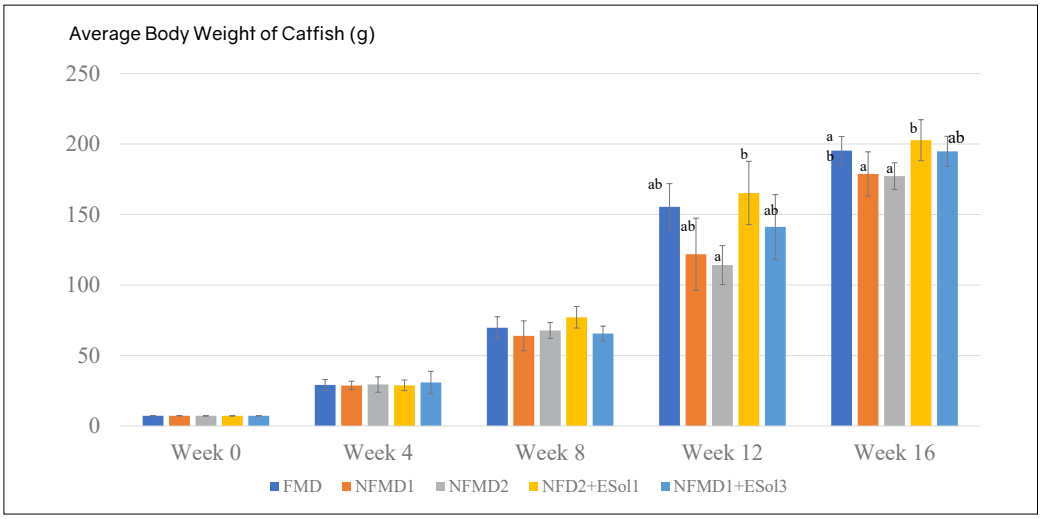

Figure 3. Growth performance of walking catfish fed with different diets for 16 weeks (P<0.05 after week 12).

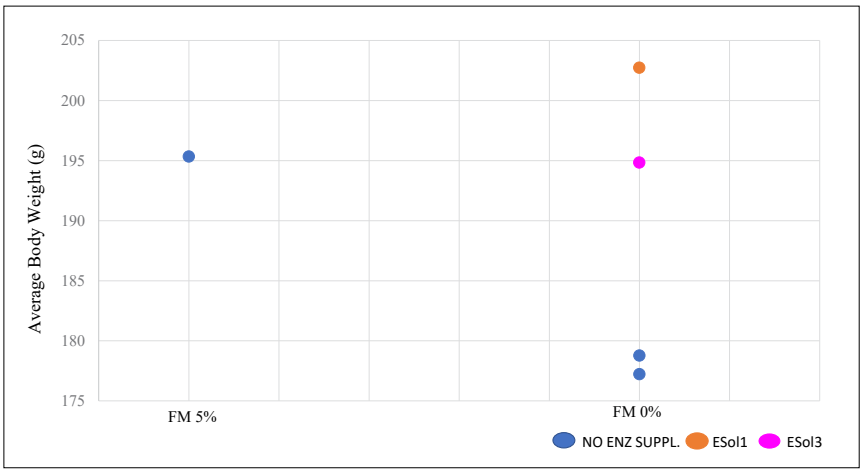

Figure 4. Average body weight of catfish comparing two factors, fishmeal inclusions (p> 0.05) and enzyme solutions (p<0.05)

Twelve weeks after initiation of the study, fish fed the ESol1 supplemented diets (Diet 4) had significantly higher average body weights (p<0.05). FCR was slightly improved in fish fed the diets supplemented with enzyme combinations. There were no significant differences among the dietary treatments for SGR (p>0.05) which averaged from 2.86 (NFMD1) and 2.97 (NFMD2).

Diet 5 (NFMD1), the 5% fishmeal replacement diet supplemented with ESol3, resulted in a 9% increase in final weight gain compared with the gain in groups with no enzyme supplementation (NFMD1). Diet 5 resulted in an improvement in weight gain to the level of FMD. Furthermore, in diet 4, the 5% fishmeal and 1.3% poultry meal replacement diet (NFMD2 + ESol1), showed an improvement in weight gain of 14.4% due to the addition of ESol1.

Summary

Supplementation of the multi-enzyme mixtures in the feeds resulted in significant improvements in growth performance in red tilapia and walking catfish. It can be concluded that the main benefit of advanced enzyme combinations is the compensation for the negative effects of replacing fishmeal with plant ingredients, and achieving better growth performance than with feeds without any enzyme supplementation. Further work is required to explore the full potential of feed enzymes on sustainable non-fishmeal diets. In conclusion, the combination of a phytase, RONOZYME® HiPhos, a xylanase, RONOZYME® WX and a protease, RONOZYME® ProAct, at an economical dosage, improves growth performance and feed efficiency of both red tilapia and walking catfish fed diets where all fishmeal has been replaced by plant or land animal proteins.

References

Dai, B., Hou, Y.B., Hou, Y. Qian, L. 2017. Effect of multienzyme complex and probiotic supplementation on the growth performance, digestive enzyme activity and gut micro organisms composition of snakehead (*Channa argus*). Aquaculture Nutrition, 25:15-25.2019. DOI: 10.1111/anu.12825

Zheng, C.C., Wu, J.W., Jin, Z.H., Ye, Z.F., Yang, S., Sun, Y.Q. and Fei, H. 2019. Exogenous enzymes as functional additive in finfish aquaculture. Aquaculture Nutrition, 00:1-12. DOI: 10.1111/anu.12995



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Effect of a nutraceutical formulation on larval and juvenile stages of whiteleg shrimp

Added to culture tanks and feed pellets, there was faster metamorphosis from zoea-2 to PL11, and a better growth performance from PL12 to PL42.

By Phuong Viet Do, Tao Tai Chau, Audric Touchet, Gaetan Gutter, Philippe Mahl and Hoang Phan

Shrimp post larvae require a high level of diverse nutrients to ensure sustainable growth and a strong immune system (Wang et al. 2015). Improving disease/stress resistance and sustainable growth performances during the post larvae stage is crucial since this is the most sensitive stage in the shrimp lifecycle (Liu et al. 2007; Xie et al. 2014). To ensure biosecurity, hatchery operators tend to decrease the use of natural food e.g. phytoplankton or zooplankton to avoid pathogenic contamination (Tacon 2017). Feed pellets are provided in small quantities for small shrimp; therefore there is a need for nutraceutical supplements to meet the nutritional requirements of shrimp (Li et al. 2017).

An earlier study at Mahidol University, Thailand, using Nutrimix (Virbac, Vietnam) - a mixture of vitamins, essential amino acids and glucuronolactone – added in feed at 0.5% w/w to feed black tiger shrimp *Penaeus monodon* weighing 5–10g showed a significant increase in the proportion of granulocytes in the haemolymph indicating that it improves the immune response (SBBU Biotec Report 2004). This article describes a study which evaluates the effect of Nutrimix on larval and juvenile stages of whiteleg shrimp *Litopenaeus vannamei* added in culture water and to the feed. It is hypothesised that Nutrimix could develop the overall health of shrimp and help the post larvae counter the adverse effects from water quality changes and other stresses incurred.

Experimental details

For the first trial, each tank with 500L water (30ppt salinity) was prepared with zoea-1 (200,000 zoea-1/m^3 density). Here, we applied Nutrimix/m^3/day for 18 days in the water at 0, 1, 3 and 5g. Following this, post larvae (PL12) were stressed with 100ppm formalin and with salinity reduction (50%, i.e. 30ppt to 15ppt) for 30 minutes. All treatments were performed in triplicate.

Figure 1. Culture tanks were set up indoors with continuous aeration during the trials.

Parameter		Trial 1: Larvae stage (g/m³/day), 18 days				Trial 2: Juvenile stage (g/kg feed/day), 30 days			
		0	1	3	5	0	5	10	15
Temp (°C)	AM	30.8±0.8	31.3±0.8	30.9±0.7	28.86±0.33	28.86±0.33	28.80±0.39	28.73±0.39	28.89±0.27
	PM	31.5±0.7	31.7±0.6	31.3±0.6	29.93±0.58	29.93±0.58	29.92±0.52	29.87±0.54	29.93±0.57
pH	AM	8.12±0.14	8.14±0.15	8.15±0.14	8.15±0.24	8.15±0.24	8.14±0.23	8.12±0.22	8.18±0.26
	PM	8.08±0.14	8.09±0.12	8.05±0.10	8.16±0.21	8.16±0.21	8.18±0.22	8.15±0.21	8.19±0.22
TAN (mg/L)		0.35±0.25	0.30±0.14	0.33±0.18	0.31±0.21	0.34±0.19	0.33±0.14	0.42±0.17	0.32±0.15
NO_2– (mg/L)		0.83±0.40	0.84±0.41	0.81±0.42	0.79±0.36	0.37±0.27	0.38±0.28	0.46±0.26	0.34±0.21
Alkalinity ($mgCaCO_3$/L)		172±11	173±12	175±12	171±11	ND	ND	ND	ND
Oxygen (mg/L)	AM	ND	ND	ND	ND	5.57±0.54	5.37±0.66	5.47±0.56	5.43±0.71
	PM	ND	ND	ND	ND	5.71±0.40	5.66±0.63	5.58±0.84	5.83±0.70

Table 1. Water parameter readings during the two shrimp trials: at larval and juvenile stages

Figure 2. Water parameters were regularly measured during the experiment

For the second trial, 1,000 post larvae (PL-12, 0.01g/PL) were stocked in a 500L water tank at 15ppt salinity. They were fed manually to satiation 4 times per day for 30 days. Feed pellets were mixed with Nutrimix. There were four inclusion rates at 0, 5, 10, 15g Nutrimix/kg feed. Water parameters during the duration of the two trials were checked regularly i.e. temperature (twice daily), pH (twice daily), total ammonia nitrogen (TAN, every 3 days), nitrite (NO_2, every 3 days) and alkalinity (every 3 days) to ensure they were in the suitable range for shrimp. All the treatments were performed in triplicate.

Effects of supplementation

Table 1 shows that water parameters were stable and in optimal ranges for all treatments throughout the duration of the two trials

Parameter	Trial 1 (g/m³/day)	0	1	3	5
Metamorphosis rate	Day #3	100% Zoea-2	100% Zoea-2	100% Zoea-2	100% Zoea-2
	Day #6	100% Mysis-2	100% Mysis-2	100% Mysis-2	100% Mysis-2
	Day #9	100% PL-1	100% PL-1	100% PL-2	100% PL-2
	Day #12	100% PL-4	100% PL-4	100% PL-5	100% PL-5
	Day #15	100% PL-7	100% PL-7	100% PL-8	100% PL-8
	Day #18	100% PL-10	100% PL-10	100% PL-11	100% PL-11
Survival rate (%) of PL-12 after 18 days		50.6±1.2^a	52.7±0.4^a	55.4±1.7^b	57.4±1.4^b
Length	Zoea-3	2.59±0.01^a	2.59±0.02^a	2.60±0.01^a	2.61±0.01^a
	Mysis-3	3.51±0.04^a	3.52±0.13^a	3.55±0.16^a	3.55±0.10^a
	PL-5	6.07±0.11^a	6.22±0.02^b	6.25±0.03^b	6.25±0.01^b
	PL-12	9.68±0.03^a	9.72±0.06^a	9.93±0.01^b	9.91±0.04^b
Survival rate (%) of PL-12 after stress	100ppm formalin	100	100	100	100
	50% salinity reduction	100	100	100	100
Values in the same row with different letter are significant different ($p<0.05$).					

Table 2. Results of Nutrimix supplemented at larval stage: Metamorphosis rate and survival rate of PL-12 during 18 days, growth in length and survival rate of PL-12 after stress challenges with formalin and salinity reduction

Parameter	Trial 2 (g/kg feed)	0	5	10	15
Weight	DWG (g/day)	0.019±0.001^a	0.020±0.001^{ab}	0.021±0.001^b	0.021±0.001^b
	SGR (%/day)	13.23±0.03^a	13.37±0.14^{ab}	13.48±0.19^{ab}	13.55±0.18^b
Length	DLG (cm/day)	0.10±0.01^a	0.10±0.01^a	0.11±0.01^a	0.11±0.01^a
	SGRL (%/day)	5.22±0.02^a	5.23±0.03^a	5.26±0.02^{ab}	5.29±0.02^b
Survival rate (%) after 30 days		90.0±1.4^a	91.6±1.5^a	92.0±1.3^a	93.1±1.5^a
Values in the same row with different letter are significantly different ($p<0.05$). DWG/DLG – daily weight/length gain; SGR/SGRL – specific growth rate/in length					

Table 3. Results of Nutrimix supplemented at juvenile stage: Growth performance and survival rate after 30 days

Figure 3. Healthy post larvae before the trial

(18 and 30 days, respectively). In Table 2, the metamorphosis index, body length and survival rates improved in the treatments with 3 or 5g Nutrimix/m^3/day after 18 days for shrimp larvae.

In the second trial, daily weight gain (DWG) and specific growth rate (SGR) of juvenile shrimp improved in the treatment diet where 15g/kg feed of the nutraceutical mix was supplemented (Table 3).

The survival rates of shrimp were recorded as >55.4% for PL12 stage supplemented with 3 or 5g Nutrimix/m^3/day, and > 93% for PL42 stage supplemented with 15g/kg feed. Shrimp at larval stage responded well to stress tests using formol and salinity reduction. All shrimp survived the challenges.

References available on request

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Disease control by hyperthermia with non-lethal heat shock

Confronted by a world of virus and bacteria, the target is to boost the innate immunity and the memory system of organisms to do the job of maintaining their homeostasis

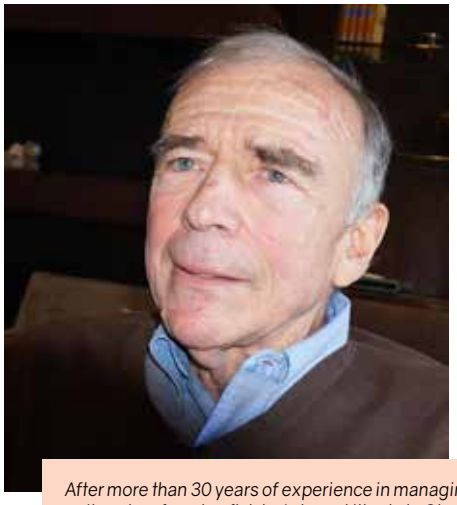
By Alain Michel

The main constraint for sustainable development of Asian seabass *Lates calcarifer* (or better known as barramundi) culture in tropical regions is still the mortality incidences encountered during juvenile production and more so, when transferring from land-based hatcheries or nurseries, to cages or ponds.

What do farmers look forward to with regards to juveniles? In my opinion, farmers look for the following:

- Juveniles with the best growth potential;
- Juveniles not infected by the local pathobiome when transferred from land-based nursery facilities to cages or ponds; and
- Juveniles at the lowest cost possible.

Therefore, mitigation measures include using juveniles coming from a selective breeding program for growth and disease resistance. In Indonesia, we ran such a program for 4 years and the methodology was well mastered using rotational mating of the same females with several males ending with a lot of families, followed by a mass intra family selection where the best of each family with DNA markers were selected. We can put juveniles in contact with the main pathogens before transfer and/or protect them by efficient vaccines, and lastly, we can have juveniles transferred at the smallest size possible to decrease the nursery costs.



*After more than 30 years of experience in managing health and disease outbreaks of marine fish in Asia and tilapia in Ghana, **Alain Michel** believes that hyperthermia is the way forward, to increase immunity at the first signs of a disease outbreak. His wish is for farm technicians to practise this approach on boosting the innate immune system to have better control of aquaculture pathogens. He has also attempted this with marine shrimp. He welcomes enquiries on heat shock treatments or any other issues in aquaculture health management.*
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What is the present situation?

Tropical fish aquaculture is still using two models. The backyard model is still predominant and popular among small farmers, but is highly unreliable. Selection is done erratically through natural selection. The production is very segmented to reduce risks and often eggs are provided by government facilities. Backyard hatcheries and nurseries operated by small farmers sell juveniles for grow-out in ponds or cages. Overall, this works well but no one single hatchery is reliable.

The sterile environment model

The modern model is based on the biosecurity concept with a production plan and which keeps the juveniles until a large size in land-based facilities to counteract the impact of disease at sea; this, however, increases the cost of juveniles. A common practice is that the hatchery starts by sanitising and removing everything in the water, with the assumption that the bacterial population which will replace the natural ones will be better. This model has reached its limits as it cannot control true pathogens and ultimately will suffer with large outbreak during the transfer of juveniles to the grow-out systems. Biosecurity is synonymous to good husbandry. It is a necessary condition but not completely foolproof and gives a kind of false insurance.

Vaccines have been introduced such as those marketed against *Streptococcus* but it is a long-term approach. Unfortunately, the market is not yet large enough to interest the major companies. Autogenous vaccines have been developed but often without real evidence of their efficacy.

New paradigm

We need to recognise that pathogens are part of our environment. We need to learn to live with them. We need to forget the illusory idea of total eradication of pathogens. Believing that "one pathogen one disease is too simple an approach." What is important is the pathobiome: the environment of the pathogen including all the other pathogens. Each local pathobiome has its own specificity which can change with time due to seasonal variations and the way of dealing with them should be adopted.

There are two kinds of pathogens which have developed different strategies:

- Opportunistic ones are always present, ready to exploit the weakness of the organisms. They should be controlled by good husbandry with regular dry out periods. They are dependent on the stocking density of the juveniles.
- True pathogens can kill more than 90% of a stock in a short period of time, even if the juveniles are very healthy . They can also infect low density culture.

Therefore, the target is to boost the innate immunity and the memory system to let the organisms do the job of maintaining their homeostasis in a world full of virus and bacteria.

Consequences

It is not necessary and useless to treat the water entering the system if we want to accustom progressively the juveniles to the pathobiome. Roughly, this can be achieved by only using a rapid

Clockwise from top, Barramundi juveniles dark black affected by iridovirus; Typical red eye is a sign of iridovirus; Scale drop disease starting after transfer in a cage and scale drop disease on barramundi.

sand filter to remove coarse detritus and a charcoal filter, if there is a fear of chemical pollution.

I am advocating to use hyperthermia and non-lethal heat shock to boost the natural defence lines of the fish. I am also advocating the use of any available effective vaccines for some true pathogenic bacteria like *Streptococcus* and to maintain at a low level the opportunistic external bacteria like *Tenacibaculum maritimum* (*T. mar*) with peroxide baths.

Experiences

All the above recommendations come from years of experience in Asian seabass farming. In the 1980s, the IFREMER centre in Tahiti developed a program on barramundi culture, initially using juveniles imported from Singapore. It then went on to achieve the reproduction of broodstock in captivity, starting with mass production of juveniles in full salinity waters and grow-out in ponds and cages.

Subsequently, there was the over 16 years involvement day and night, over six months per year in the set-up and the development of an ambitious production project of several species in offshore cages off a small island in the Seribu archipelago in the Java Sea north of Jakarta. There was a special emphasis on barramundi production. More recently, there is work done in Sri Lanka on a barramundi farm operating in offshore cages starting with fry imported from Australian hatcheries. We shifted to captive broodstock by selection of the best fish from the cages. Since early 2018, all the new juveniles entering the cages came from local spawning.

There were also discussions on disease management. At a Darwin hatchery in Northern Territory, Australia, a close friend Jerome Bosmans provided the juveniles for a Marine Harvest development project in Tiwi Island. Discussions were also held at Singaporean and Indonesian farms, namely Barramundi Asia, Indomarine, and Marine Life Aquaculture. There were also discussions and inputs of the MSD team in Singapore and with Susan Kueh Gibson in Perth University, Australia.

In short-term trials, at the Australis hatchery in Vietnam, we worked on heat shock treatment which Josh Goldman, co-founder and CEO recognised as effective, which he supported with an email message, "I am boiling the fish and it works." There were experiments on heat shock treatments on invertebrates (shrimp and sea cucumbers), on the tilapia in Sumatra (Indonesia) and Ghana, and on frogs in France.

From all these field experiences, the data collected and the results obtained so far, I am more and more convinced that non-lethal heat shock treatment to boost anti-stress biological pathways can mitigate most of the disease problems. It is a treatment dealing with the strains of pathogens in real time which will integrate virus mutation or even a change in serotypes for bacteria. It acts at the cell level with intra and extracellular effects. Heat shock proteins have been conserved through evolution from bacteria to humans and their role is pivotal.

Disease in tropical fish culture

Most of the viruses and some bacteria can cause true diseases. In the case of the barramundi the list is long.

- Four viruses: nervous necrosis virus (VNN), irido infectious spleen and kidney necrosis virus (ISKNV), scale drop disease virus and herpes virus;
- Two bacteria: intracellular *Vibrio* for big belly syndrome, and *Streptococcus iniae*. *T. mar* has an intermediate position and is more opportunistic, but is susceptible to spread rapidly under some conditions.
- Among the true opportunistic bacteria, there are mainly different species of *Vibrio* such as *Vibrio alginolyticus*, *V. harveyi* and others.

With regards to parasites, there are:

- flat worm Benedenia which is very harmful in cages if not treated before fish reach 300g
- *Cryptocarion irridans* which often infects broodstock
- Amylodinium which often infects larvae

Today, PCR diagnostics are available for almost all the viruses and bacteria. Therefore, we are also finding the same diseases in other marine fish species such as in the grouper *Epinephelus* spp, snappers *Lutianus* spp, pompano *Trachinotus blotchii* and cobia *Rachycentron canadum*. Unfortunately, in aquaculture there is always the possibility of new emerging diseases which are occurring in our high density culture systems which are interesting playgrounds for disease outbreaks.

Barramundi schooling behaviour enhances spread of diseases.

Barramundi mass mortality after transfer

Left: Juvenile barramundi affected by iridovirus becoming dark black.

Right: Pale gills found both in iridovirus and scale drop disease infected fish

Science

Today, science has often emphasised on biosecurity and more biosecurity. This will help in the eradication of all the pathogens and the use of antibiotics will be the last resort when not knowing what else to do. There are many kinds of products from immunostimulants, probiotics, prebiotics and functional feeds which have been advocated to mitigate health issues in fish farming. However, in my opinion, there are still some questions on their efficacy.

Hyperthermia upregulating the heat shock proteins

In the presence of pathogens, hyperthermia can up-regulate the production of heat shock proteins. It came from a serendipity effect on VNN followed by an extension to big belly disease to replace the use of high doses of antibiotics. At the same moment there were results published on host-virus relations and temperature. There was also a long series of trial and error experiments to solve the emerging iridovirus mortality after obtaining negative results with immunostimulant and specific vaccines.

The first experiment was carried out on fish brought back to land-based facilities from an affected cage. The preliminary results were positive. After using long duration treatment, we progressively decided on the protocol which we advocate now: non-lethal heat shock of short duration, 30 minutes repeated on a weekly basis till the mortality is stopped. This has been confirmed by recent experiments demonstrating the up regulation of the heat shock lasting 24 hours.

Applying non-lethal heat shock treatments

Up to now, this was used in land-based facilities following disease events and reacting to the first clinical signs. It was done with industrial boilers or steam boilers. The water level is lowered in the tank to be treated. Water is heated at 55°C and an equal volume of water in the treated tank is replaced with the heated water, which is enough to increase the temperature in 30 minutes from 29°C to 39°C. The temperature is maintained at this level for 30 minutes. Then the water replacement is resumed.

A variant is to heat directly the tank with steam injected in an airlift placed in the middle of the tank. This decreases the risks of burning the fish with a very progressive increase of temperature.

The target now is to be able to reduce the necessary time in the nursery tanks for productivity and cost reasons. I have begun to run a series of trials to do the heat shock treatments directly in the cages, using tarpaulin and a barge with a steam boiler and a movable airlift system for steam injection plus a generator and a blower. Still many improvements are required.

“ A combination of these non-lethal heat shocks with efficient vaccines commercially available, and regular peroxide baths to maintain a low level of the opportunistic bacteria, is presently the optimal protocol to mitigate disease problems. ”

Hyperthermia approaches

Our field results obtained so far with the non-lethal heat shocks or by manipulating temperature allow us to say that the hyperthermia approach has been applied successfully on barramundi:

- VNN when it hits a larval batch after two weeks. However, if VNN is present at spawning or appears at the first week, the recommendation is to discard the affected tanks;
- iridovirus;
- big belly bacteria;
- scale drop disease virus or herpesvirus.

In general, the hyperthermia approaches seem to be effective on viruses with a vaccination effect. The exact mechanism of action of the biological pathways of up-regulation by hyperthermia is not fully understood. Sometime there are contradictory effects revealing a dual role linked with their internal and external effects. There is a need for other experiments to be done especially on *Streptococcus* following some positive answers.

A combination of these non-lethal heat shocks with efficient vaccines commercially available and regular peroxide baths to maintain a low level of the opportunistic bacteria is presently the optimal protocol to mitigate the disease problems.

The next step

Often, there are questions concerning the right timing of the treatment. Some recent results on tilapia-iridovirus demonstrate the efficacy of the treatment on small fry of around 0.5g, thus opening the way to mass treatment at a low cost. I have demonstrated that it is possible to introduce the pathogens kept in frozen conditions in deep freezers in the nursery tanks to induce the disease to be treated immediately to obtain the protection by the vaccination-like effect.

The question is: could it be possible to induce the infection of several pathogens at the same moment and to obtain a poly protection like a polyvalent vaccine? It appears that the innate immune system can learn very efficiently at the early stages. However, it is very important to note that non-lethal heat shock treatment is not preventive.

Updates on EHP and shrimp disease threats

EHP has become the major threat and some measures to contain the spread are proposed

At Infofish Shrimp 2019, held from 12-14 November, there was a panel on "Updates on shrimp disease threats". Three presenters discussed recent knowledge: **Dr Loc Tran**, ShrimpVet Lab, Minh Phu AquaMekong, Vietnam; **Dr Kallaya Sritunyalucksana-Dangtip**, National Center for Genetic Engineering and Biotechnology (BIOTEC), National Science and Technology Development Agency, Thailand and **Dr Jumroensri Thawonsuwan**, from the Songkhla Aquatic Animal Health Research Center (SAAHRC), Department of Fisheries, Thailand (DOF). Jumroensri gave results from the surveillance conducted by DOF Thailand as well as some nationwide measures to reduce pathogens in shrimp farming.

Both Loc Tran and Jumroensri discussed threats from several diseases but in this report, the focus is on *Enterocytozoon hepatopenaei* (EHP) which Kallaya said, "Overall, infections have reached epidemic proportions in Asian shrimp aquaculture. Specifically, in Thailand, there is a high prevalence of EHP at 60.1% of ponds affected." Jumroensri showed that in Thailand, DOF focuses on detecting disease pathogens in samples. In the surveillance in 2019, the presence in post larvae and grow-out was: EHP >Vp_{AHPND}>WSSV>IHHNV>YHV. The presence of EHP have increased to 26.5% in 2019, from 25.1% in 2017 and 17.5% in 2018 whereas detections of *Vibrio parahaemolyticus* (Vp_{AHPND}) went down to 2.1% in 2019 from 3.6% in 2018 and 6.4% in 2017. Furthermore, EHP detections were highest in *Penaeus vannamei* juveniles at almost double that in post larvae.

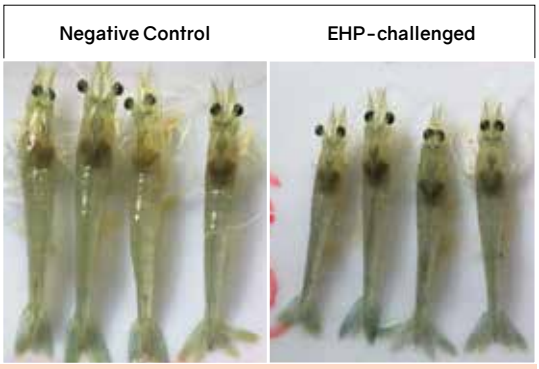
Loc Tran who is Founder- Director of ShrimpVet reviewed lessons learnt from the emergence of three disease threats affecting productivity and margins for Asia's shrimp industry. These are the early mortality syndrome/acute hepatopancreatic necrosis disease (EMS/AHPND), EHP and white feces disease (WFD). Besides these, there are of course challenges from white spot syndrome virus (WSSV), shrimp haemocyte iridescent virus (SHIV) as well as the antibiotic residues, as farms resort to quick remedies. Based on samples tested at ShrimpVet Laboratory, Loc Tran showed an increasing trend in PCR positive results for EHP. In 2019, EHP was positive in 60.3% of grow-out shrimp samples versus 43% in 2018. In contrast, it was only 33% for the AHPND pathogen in 2019. He said, "There is an accumulation of this pathogen in farms and incoming water and living with EHP is becoming a norm."

EHP

Kallaya who heads the Aquatic Animal Health Research Team (AQHT) at BIOTEC gave updates from research done at BIOTEC and in collaboration with experts on microsporidia at CEFAS and University of Exeter, UK. EHP is a microsporidia related to fungi and infected shrimp grows slowly. However, growth retardation is not clearly evident before two months of culture. The research team seeks to answer several questions listed below. Answers will help them design possible control strategies.

Transmission route of EHP and its mechanism of infection

"For the first time, we have showed that EHP is transmitted horizontally when we added infected shrimp to a tank of naïve shrimp. At the laboratory, infection occurred within 14 days. This is through faeces of the infected shrimp which contained the spores. It has taken us a long time to purify the spore for a better understanding of the infection mechanism. The EHP spore germination is an important process for EHP infection in shrimp. Recently, we can purify the EHP spores and develop the assay to induce spore germination via



Growth retardation in shrimp when challenged with EHP. Photo from Loc Tran, ShrimpVet Lab, Minh Phu AquaMekong.

The panel was chaired by Oscar Hennig, Benchmark Holdings (centre right). Members of the panel were, from left, Dr Anil Kumar, MPEDA, India; Loc Tran; Kallaya Sritunyalucksana-Dangtip; Jumroensri Thawonsuwan and Dr Coco Kokartin, Ministry of Marine Affairs and Fisheries, Indonesia.

polar tube extrusion. The assay allows us to test chemicals and disinfectants that can be used to inhibit the germination process in the hatchery and grow-out pond (Aldama-Cano et al., 2018)," said Kallaya. Figure 1 shows the germinated EHP spores (active spores) and the non-germinated spores (inactive spores).

What causes shrimp to grow slowly

With the purification of the spore, Kallaya said that the team then went on to sequence the genome of the microsporidian (Wiredu Boakye, et al., 2017). "The genome showed missing genes encoding for the enzymes involved in the glycolytic pathway in EHP. As glycolysis is the fundamental pathway of ATP generation in eukaryotes, when the genes of the pathway are lost, energy for EHP growth is obtained directly from the shrimp resulting in shrimp slow growth."

New PCR method

Arising from the work on the EHP genome, BIOTEC and Centex Shrimp has developed a method (SWP-PCR) to detect EHP. It is recommended to use for detection of EHP in non-shrimp samples (faeces, feed and environmental samples) for potential EHP carriers (Jaroenlak et al., 2016). The new SWP-PCR method did not produce false positive results from closely related microsporidia whereas the existing PCR detection methods (SSU-PCR) can give false positive test results due to cross reactivity of the SSU-PCR primers with DNA from closely related microsporidia that infect other aquatic organisms.

Environment reservoir of EHP

"Despite that SWP-PCR methods has been established and that we have a better understanding of the mechanism of EHP infection, the prevalence of EHP is still high in our last survey in Thailand. We hypothesize that there might be the environmental reservoir(s) of EHP in the pond that has been overlooked. Our on-going task is to use plankton nets with different mesh sizes to filter the water collected from the EHP heavily infected ponds. By using metagenomics approach, we hope to find the reservoir(s) of EHP in the pond, if there are any," added Kallaya.

WFD–EHP: A perfect combo

Loc Tran gave this description when the shrimp is first infected by EHP. WFD is bacterial linked and the result of sloughing of microvilli and cells of the hepatopancreas. *Vibrio* has been isolated from WFD infected animals. In some laboratory trials, he showed that a pre-infection with EHP may increase the severity of WFD in a *Vibrio* challenge. Similar phenomenon is observed in the field leading to two types of WFD, a treatable one when there is *vibriosis* only and an untreatable vibriosis + EHP.

Some practical solutions specifically for EHP

From the current molecular understanding, Kallaya gave some practical suggestions on the control the EHP including.

- Check the post larvae with PCR-SWP before purchase.
- Laboratory trials showed a complete inhibition of spore germination occurred when the spores were frozen at –20 °C for at least 2 hours. The chemicals that yielded 100% inhibition were 15 ppm $KMnO_4$ (potassium permanganate) for 15 minutes, 40 ppm of 65% active chlorine for 15 minutes or 10 ppm of 65% active chlorine for 24 hours and 20% ethanol for 15 minutes (Aldama-Cano et al, 2018). There was no loss of fatty acids profile of frozen polychaetes after 3 months.
- Spore germination occurs at high pH (≥9). Pre-treatment of grow-out ponds by increasing pH before stocking might help to induce spore that are contaminated in the pond from the previous crop to germinate. Germination without the host will cause the spore to be inactivated and finally die.

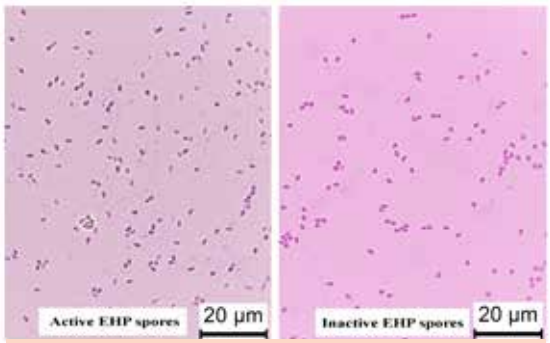


Figure 1 Germinated EHP spores (active spores) and the non-germinated spores (inactive spores). Photos from Kallaya Sritunyalucksana-Dangtip, BIOTEC.

According to Loc Tran, it is important to control EHP at the hatchery level. He recommends PCR checks of all inputs (brood stock, brood stock feed) and importantly to freeze live polychaetes. "This is a very effective control, but the downside is reduction in fecundity which brings up nauplii production costs by 30%. The total cost implication is small; at the hatchery, nauplii production cost is only 10% of total production cost and for the farmer, the post larvae cost only account for 5% of total farming costs. Therefore, the hatchery owner and farmer need to collaborate and share costs to prevent EHP in the farming system."

Nationwide control on pathogens

Over in Thailand, Jumroensri who is Director, SAAHRC said that the DOF has set up several measures and legislations to reduce disease impacts in hatchery and nursery stages. There is the mandatory standard for pathogen-free *P. vannamei* nauplii production system where compliant farms are accredited. Today there are 50 farms accredited. The "cleaning up project" for marine shrimp hatchery and nursery using real time PCR is for eight diseases (WSSV, IHHNV, YHV, TSV, IMNV, Vp_{AHPND}, EHP, SHIV) and in this 245 farms are in the white list where DOF does the "Lot by Lot project" where these eight pathogens are tested in post larvae before stocking in grow out ponds.

In terms of control strategies DOF works with BIOTEC- once information is obtained, some protocols are developed for the transfer to farmers. One example is on pond treatments, using fertilizers and increasing alkalinity to induce spore shooting. The DOF has a *Bacillus* probiotics and herb extract (galangal extract) to reduce infections.

References

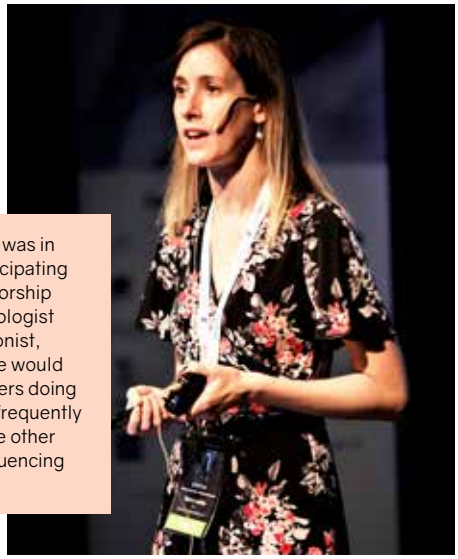
Aldama-Cano, Diva & Sanguanrut, Piyachat & Munkongwongsiri, Natthinee & Ibarra-Gamez, Jose & Itsathitphaisarn, Orn & Vanichviriyakit, Rapeepun & Flegel, Tim & Sritunyalucksana, Kallaya & Thitamadee, Siripong. (2018). Bioassay for spore polar tube extrusion of shrimp *Enterocytozoon hepatopenaei* (EHP). Aquaculture. 490. 10.1016/j.aquaculture.2018.02.039.

Wiredu Boakye, Dominic & Jaroenlak, Pattana & Prachumwat, Anuphap & Williams, Tom & Bateman, Kelly & Itsathitphaisarn, Orn & Sritunyalucksana, Kallaya & Paszkiewicz, Konrad & Moore, Karen & Stentiford, Grant & Williams, Bryony. (2017). Decay of the glycolytic pathway and adaptation to intranuclear parasitism within Enterocytozoonidae microsporidia. Environmental Microbiology. 19. 10.1111/1462-2920.13734.

Jaroenlak P, Sanguanrut P, Williams BA, Stentiford GD, Flegel TW, Sritunyalucksana K and Itsathitphaisa O (2016). A Nested PCR Assay to Avoid False Positive Detection of the Microsporidian *Enterocytozoon hepatopenaei* (EHP) in Environmental Samples in Shrimp Farms. PLoS One. 2016;11(11):e0166320. Published 2016 Nov 10. doi:10.1371/journal.pone.0166320

Bioengineered capture proteins for easy-to-interpret detection of disease targets

Gaskiya Diagnostics has a platform technology using bioengineered capture proteins to bind target analytes with a high degree of sensitivity and specificity. The capture proteins can bind peptides, small molecules and nucleic acids, thus facilitating the detection of a variety of disease types. An array of sample types can be tested, and small to large sample volumes accommodated. The bioengineered capture proteins are thermally stable, and yield consistent and easy-to-interpret detection of disease targets. They are being incorporated into user-friendly, field-use diagnostic tests.



Dr Mary Larkin was in Singapore participating in Hatch's mentorship program. Toxicologist cum fish nutritionist, Larkin said, "We would like to see farmers doing diagnostics as frequently as they measure other parameters influencing health."

In 2019, Dr Mary Larkin was in Singapore participating in Hatch's mentorship program. Toxicologist cum fish nutritionist, Larkin is co-founder and CEO at the startup Gaskiya Diagnostics, based in Maryland, USA. The startup is developing innovative rapid, low-cost, and easy-to-use diagnostic tests for aquaculture. Biomolecular Engineer Dr Hadley Sikes, co-founder and Senior Scientific Advisor, is the inventor of the technology at Massachusetts Institute of Technology (MIT) and has a partner laboratory at the National University of Singapore (NUS). Co-founder and COO, Dr Scott Gaboury is a materials chemist. Chris Moad of Early Charm, LLC, directs business development.

Initially, Gaskiya (which means truth in a Nigerian language) applied the platform technology to developing simple-to-use and low-cost diagnostics for human disease agents that plague the developing world. At the Hatch office, Larkin explained, "We are now applying the technology to developing tests for aquaculture. There is a great need for user-friendly tests that can be used on-site. We are estimating the price to be in the range of USD5/test, substantially lower than most products in the market today. Furthermore, it is fast with results within 10 minutes and the farmers can perform the tests themselves. Currently, the only option available to most farmers is to take samples, send them off to labs at significant expense, and then wait days to weeks for results. We want to put products into their hands that enable them to diagnose and manage disease in real-time and also save them money."

The innovation is unique in that Gaskiya's bioengineered capture proteins specific to a disease target are incorporated into multi-layer, paper-based test strips. A test sample undergoes various reactions as it flows through the paper layers, ultimately yielding an easy-to-read colorimetric result. Positive tests initiate the formation of a stable hydrogel, the colour of which can be related to a concentration of the disease-causing agent. No expertise and no additional equipment are required to perform the test. Farmers can perform these tests on-site and obtain meaningful results that help them manage disease and promote health. A signal amplification reaction step built into the platform enables detection of even very low concentrations of the target. Larkin explained, "For the fish or shrimp farmer, those test results can help the farmer decide what actions to take in terms of treatment or if he/she should rapidly harvest before the whole stock is lost."

Gaskiya's capture protein offers distinct advantages over antibodies incorporated into similar rapid diagnostics tests. "They are inherently thermally stable because their core structure comes from a thermophile," added Larkin. This is an important feature for a test being used on-site in hot climates. "Also, unlike antibodies, we do not need an immunogenic target. We can capture a much greater range of targets than antibody-based tests."

The team at Gaskiya Diagnostics is committed to keeping the test costs as low as possible to facilitate more frequent water testing by the farmers. "For water quality monitoring, farmers take samples daily. We would like to see farmers doing diagnostics as frequently as they measure other parameters influencing health."

In terms of target pathogens, Gaskiya is focusing on streptococcal disease in tilapia where in China alone, losses are USD1.5 billion annually. In the business model, Gaskiya will work with feed and vaccine producers, and pharmaceutical companies. The market to combat disease outbreaks is large and Larkin said, "As a company we will benefit by adding value to these large companies, and as they grow, we will monetise."

To date, Gaskiya has the support of several leading institutions and also Tropo farms, producer of Volta Catch tilapia in Ghana, and BASF. It is seeking USD310,000 in funding to move to the next step of field testing and sales. www.gaskiyadiagnostics.com

Oral vaccines platform against *Streptococcus* in the tilapia

Argentina-based startup, **FeedVAX**, is developing a platform of oral vaccines which are feed-based, safe for the environment and are effective vaccines that can protect farmed fish from disease and replace the use of antibiotics in aquaculture. The 4-member team led by CEO Luis Barletta began developing oral vaccines for the tilapia 2 years ago. As a cattle breeder, Barletta had experienced the "pain" that the vaccination process gave producers; each step of the process increased the stress of the animals, as well as that of the employees. Given this, he went on to seek a way to simplify the vaccination process and at the same time minimise the risks associated with vaccination.

He said that aquaculture presents a much more serious problem. Thus, the company's mission is to provide customers with an affordable and efficient oral vaccine to protect farmed fish. On why the tilapia, Barletta said, "In the market, there are no oral vaccines. That is why we are here with oral vaccines. Our other choice was to work on oral vaccines for the salmon but there is less work being done on the tilapia, which also has a much shorter life cycle."

*CEO **Luis Barletta** leads a 4-member team to develop oral vaccines for the tilapia.*

Barletta and Verónica Romero Brancato, a biochemist, are part of the entrepreneurial team. They have on their team, biochemist Dr Max Wilda with experience in DNA vaccines platform for the salmon. "Today we are working on oral vaccines for *Streptococcus* for the 400,000 tonnes per year Brazilian tilapia industry. We already have letters of intent to supply a total of 35 million doses to Geneseas and Aquabel, two of the largest tilapia farms in Brazil.

At the moment, Feedvax is working with different compositions of carriers for this oral vaccine, said Brancato during a discussion at the Hatch Blue's office in Singapore in November 2019. "Our oral vaccines are coated onto feeds post pelleting. We are approaching *Streptococcus* infections with two tools: vaccines and a cocktail of phages. So, on one hand we will have the prevention and on the other, in case of stress, we can use a booster vaccine or use the cocktail of phages. In both cases, the aim is to avoid use of antibiotics," added Barletta.

One of the challenges with vaccines is in determining the efficacy of each vaccination. "One of our aims is to ensure that the fish will be protected throughout the growth cycle. The team is still at the early stages in its vaccine development and will conduct tests at the laboratory level on efficacy and at the farm level in a few months. "

On the types of *Streptococcus* bacteria used for vaccine development, Brancato said that they are working on every type of *Streptococcus* found globally, even though some strains are not in the tilapia in Brazil. "Part of our development is using algorithms to build synthetic antigens that are immunologically stronger than natural based ones. We designed the vaccine to be used at the very early fry stages, where it is impossible to use the injection mode of application common with current vaccines available in the market. Also, we are focussing on highly conserved antigens present in most serotypes, which will not undergo selective pressure in a matter of a few generations."

Barletta said, "For us, it is important that we present a simple solution for the farmers. This approach with oral vaccines is simple from an operational aspect. It is not just about efficiency." He acknowledged that it is difficult being a farmer. www.feedvax.com

Marketing Philippines shrimp globally

Some perspectives to market shrimp by Chingling Tanco at the 12th Philippines Shrimp Congress 2019

By Norlyana Mohamad Termidzi

The global shrimp industry has seen significant changes within the last 10 years with early mortality syndrome (EMS) outbreaks in Asia since 2010 causing supply disruptions. On how shrimp ex-farm prices in the Philippines fared during the last 10 years, **Chingling Tanco**, Managing Director, MidaTrade Ventures International Inc/MidaFood Distributors Inc referred to her presentation at PhilShrimp 2017. She said as supply was down in 2014, farmgate prices were up to PHP220/kg (USD4.91/kg, base 10g) and as supply increased again in 2016, prices were down to PHP120/kg (USD2.68/kg). In the Philippines, prices are quoted at size 100/kg or 10g each.

In 2018, new trade flows emerged within the shrimp trade originating from Ecuador and India, which resulted in an increase of shrimp exports in terms of volume to the US, Europe and China. "But here in the Philippines, we only started exporting in 2013 when the world supply tumbled because EMS reached Thailand. If we look at the price trend today, in 2019, it is the lowest since the last five years we have been exporting." (Figure 1).

Chingling Tanco is Chair, Fisheries & Aquaculture Board and Managing Director, MidaTrade Ventures International Inc/ MidaFood Distributors Inc. Chingling is active in the shrimp industry in the Philippines and was among the proponents in bringing vannamei shrimp farming into the country. She has strongly advocated that seafood processors upgrade and expand capacity to reach export markets. This message is reflected in her presentation titled "USA & European Markets Plus Maybe A Few More Bonus Issues" at the 12th Philippines Shrimp Congress (PhilShrimp) held in Bacolod City, 20-22 November 2019.

Most of the vannamei shrimp production, estimated at 60,000 tonnes in 2019 is for local consumption where prices are much better than exports, especially during the colder months of December to March where stocking is less due to fears of white spot syndrome virus (WSSV). In 2019, the lowest farmgate price for 15g shrimp was PHP150/kg (USD2.96/kg). The November price was higher at PHP220/kg (USD4.35/kg) and was expected to rise to PHP350/kg (USD6.90/kg) over the peak Christmas season.

In 2019, the Philippines exported USD40.6 million of shrimp products, mainly to 10 markets led by Japan, USA, Korea, France and Taiwan. In 2018, Japan imported 2,452 tonnes of frozen shrimp from the Philippines. The product range from the Philippines include frozen-head-on individually quick freezing (IQF), block frozen and peeled. Chilled products are presented as head-on shell-on (HOSO), according to the Export Marketing Bureau Services, Department of Trade and Industry.

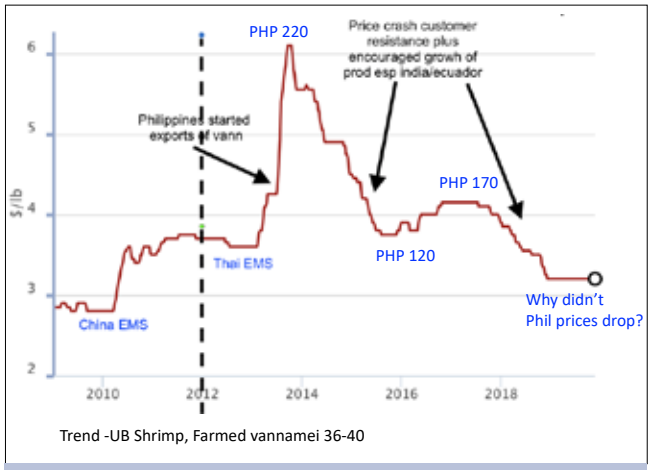


Figure 1. Urner Barry price trend for Asian White 36-40 amidst EMS. Note how the prices in the Philippines reflected the demand and supply situation worldwide with the exception of 2018, where local prices remained resilient despite high supply.

Perspectives on markets and potential for Philippines shrimp

US market

This is one big, homogeneous and transparent commodity market, which is easy to penetrate. In 2019, total shrimp imports into the US was 696,405 tonnes. Retail makes up about 40% of the market and their shrimp are usually peeled (from India and Indonesia), EZPeel or Zipperback (from Indonesia) or cooked (from Thailand, India and Indonesia). The remaining 60% of the market is food service with the shrimp either peeled or headless shell-on (HLSO). When exporting to the US, suppliers can expect big orders and long-term contracts with 100 to 200 containers of shrimp to be shipped over between 3-6 months. There is a preference for 1-3 shrimp sizes in order to fulfil these large orders faster and more efficiently. Producing companies can set up their own distribution operations in the USA, as is done by India and Thailand.

The main supplier for the US is India with its newest big plants of 40 to 100 tonnes/day capacity. Many seafood companies have several plants across India to tap into the country's vast shrimp resources. It is observed that India has dramatically increased its share in the global shrimp market producing large volumes at low prices (Rubel et al., 2019). Indian shrimp exports are either IQF, peeled, deveined, tail-on (IQF PDTO) or IQF cooked or block frozen headless (treated). Besides India, Indonesia also supplies the US with their peeled big-sized and small-sized shrimp. Despite having reduced the number of plants from around 40 to 20, Indonesian plants may have a minimum volume of shrimp production at 15 tonnes/day whereas capacity of the largest plant could go up to 80 to 100 tonnes/day.

"The US is not a good market for the Philippines because the US commodity game is played by Philippines companies with basic contact freezers without much choice for the other markets. Therefore, Mida chose a niche market for their suppliers. There are only a few with IQF capacity giving them better product form options; some suppliers are able to produce IQF products but are without third-party certifications which make it difficult to export to the US. To supply most large US players, the 'entrance ticket' into the market requires the processing plant to have at least 1-star BAP (Best Aquaculture Practices) and quickly moving to 3-4 stars," said Chingling.

Clockwise from top left:Philippines shrimp; A preference is cooked shrimp with colour 27-30 on the colour fan; for the Chinese market, dark raw and cooked shrimp from Thailand and dark, almost black colour (A5) raw shrimp.

EU markets

As it is made up of several different countries, the European Union (EU) market is diverse, non-homogeneous and difficult to penetrate due to tighter and different restrictions, compared to the US. Imports are estimated at more than 900,000 tonnes in 2019 (Sackton, 2020). Product labels must be written in multiple languages. "Unlike the US, EU countries require more certifications from the producers as concerns on animal welfare are being included in the handling requirements. It is a difficult market to enter but once qualified, it is a good value market," said Chingling.

"Some of the additional certifications are the Business Social Compliance Initiative (BSCI). Consumers prefer Aquaculture Stewardship Council (ASC)-certified products because they are already familiar with the Marine Stewardship Council (MSC) for their wild caught seafood. The EU has more intensive third-party audits and multiple audits down the value chain to ensure compliance."

The diversity of the EU market means varying product forms for each country. In Spain and France, there is a large market for head-on raw material. Besides that, they prefer good colour shrimp and have a low tolerance for burst hepatopancreas. Spain and France together account for 70,000 tonnes of the EU market and most of that is supplied by Ecuador. Germany requires little to zero chemical treatment of the shrimp. Italy opts for the high glazing in shrimp and has confusing declarations of weight. Chingling said, "We see a bag there that says 70% but it's really 65% or higher glaze than what they actually declare."

The UK, on the other hand, prefers small-sized shrimp, part of the high-end peeled market, which requires certifications as well as numerous validations for certifications and process audits. Some of the imported shrimp are sold in supermarkets such as Tesco, Waitrose and Sainsbury's. "To keep up with the huge demand for small shrimp in the UK, Mida Food Distributors Inc. is working with some growers to just produce 10-12g ASC certified shrimp and the market for that alone is about 6,000 tonnes/year," said Chingling.

The UK's oriental market takes cheaper commodity peeled shrimp which is dominated by shrimp from India. At the other end is "super premium product" which is an attractive market selling fresh in local supermarkets, ready to eat at £20/kg. "This is a strong market but you need to be able to comply. We have been working on the certification for two plants in Indonesia since 1.5 years ago, and are only halfway there," said Chingling which explains the difficulties with the certification process.

China market

The Chinese market was also discussed. Based on the data gathered by Anderson et al., at GOAL 2019, China imports as much as 700,000 tonnes of shrimp per year and it is fast becoming the largest shrimp importer in the world. Major retailers are requiring third-party certifications before importing the shrimp into China. Ecuador is China's supplier for HOSO shrimp, while peeled shrimp are from India and Indonesia. "The Ecuadorian shrimp have strong hepatopancreas which does not burst as they come from extensive ponds. The main sizes are 30-40g and smaller ones are 25g or less. As for shrimp from Saudi Arabia, it is possibly the same as it is also farmed in extensive systems with yields of 40 tonnes/10ha pond; the shrimp are also 25g or less," said Chingling.

The imported HOSO large-sized shrimp for China are either frozen from live harvest, raw or cooked, and dark coloured which can give a premium on the price at USD1/kg or more. There is a difference of between US80 cents to USD1/kg between live and chilled harvest. In terms of colour, the preference for cooked shrimp is between 27-30 on the colour fan. Other competitors supplying large shrimp are Thailand (size 20-30/kg) and Vietnam. The latter's shrimp can be up to 33g and more. There is also some competition from India because it had just started HOSO shrimp from live harvest. Peeled shrimp comes from India and Indonesia.

Shrimp processing in the Philippines

There are 29 seafood processing plants in operation in the Philippines. Philippine exporters continue to renovate their respective plants and install new equipment like tunnel freezers but still maintain the reliable contact freezers. New processing plants have been built with the latest equipment and larger cold storage in the past 5 years (Pio, 2019).

Despite this, Chingling said, "Processors continue to face some challenges in the transition from processing monodon shrimp to vannamei shrimp products, which will take some time to overcome. Many are equipped for IQF but only two have cookers. Only five have the approval to export to EU markets. Therefore, investors such as Fisher Farm, Dataj and SAFI invest in equipment and plant upgrades. Besides that, good production leaders, marketing and purchasing are also essential to ensure a successful operation. The plants must be installed with the latest systems for traceability and good production."

On product forms, she added that before it was just HOSO monodon shrimp to Japan. With the vannamei shrimp, product forms are many: HOSO, HLSO, peeled, tail-on, tail-off, raw or cooked, treated or not with either phosphates, metabisulfites

or non-phosphates or variations of glazing. Then, there is the need for certifications for food safety, environmental and social responsibility and ethical audits. "To date, the Philippines only has two plants with BAP certification and the world is already moving on to animal welfare, so they must follow the trend and obtain the necessary certifications to compete with the other exporting countries. In animal welfare, it requires non eyestalk ablation in hatcheries, and products need to be certified that there is no poultry meal and that a reduced amount of fishmeal is used in feeds."

> **" Being certified may end up being costly and challenging at first; however you will reap long-term benefits from getting certifications when you wish to compete with the other major exporting countries to penetrate your desired market. "**
> **- Chingling Tanco**

What markets demand

When it comes to supplying and exporting seafood products to the US and the EU, exporters are required to abide by BAP standards and hold various certifications, such as the ASC and GlobalGAP to ensure that the products are ethically prepared and sustainably sourced. In the Philippines, companies are trying to understand and obtain the ASC certification; therefore, most companies are without the ASC certification while some processing plants do not even have the Hazard Analysis and Critical Control Points (HACCP) certification.

The EU market is more towards ASC and GlobalGAP which are more difficult to attain because of the requirement for Biodiversity Environment Impact Assessment. In the UK, retailers respond more to the market; they are serious on environmental and social responsibility, so audits are even more difficult," said Chingling adding, "Our competitor here is Vietnam. I think with government support in Vietnam, almost all the plants have ASC certifications."

Over the past 8 years, Mida has been working with the Philippines shrimp farming and processing segments as plants get ready. It has assisted plants in Bacolod, Cebu, Luzon and General Santos to access markets in the US, Europe and Mauritius. In March 2018, Mida started processing with two plants in Indonesia and by the first quarter in 2020, an initial plant audit is expected. This will be by a UK processor which processes cooked, chilled ready-to-eat shrimp with a shelf life of 10 days for supermarkets such as Sainsbury's and Tesco. The reward for selected plants is a stable and good price for as much as 1,000-2,000 tonnes per year of product. But, for this processor, the preference is supply from an integrated operation with 4-star BAP and ASC certification. Their end-user supermarket Sainsbury's will also do a plant and facility audit.

Chingling's message was, "To develop markets for the Philippines shrimp, we need to make sure that the processors are up-to-date, their production personnel are flexible and nimble and the plants have achieved the required certifications to produce as well as export products. Being certified may end up being costly and challenging at first; however you will reap long-term benefits from getting certifications when you wish to compete with the other major exporting countries to penetrate your desired market."

Reference
A Strategic Approach to Sustainable Shrimp Production in Indonesia 2019.
https://media-publications.bcg.com/BCG-A-Strategic-Approach-to-Sustainable-Shrimp-Production-Thailand-July-2019.pdf

Giant Prawn 2019

Bringing up giant freshwater prawn production in Asia to new levels with biotechnology and genomics

Participants at a post conference visit to a freshwater prawn farm in Yangzhou city, Jiangsu province.

The Giant Prawn Conference Fifth Edition, jointly organised by the Asian Institute of Technology (AIT), Thailand and Shanghai Ocean University (SHOU), China, was held for the first time in China from 15-18 November, 2019. A WhatsApp chat group was also formed to promote further discussion on giant prawn culture. The event included a display area featuring companies promoting feeds and additives to boost aquatic animal health, aquaculture programmes and training available at AIT and SHOU, as well as publications highlighting research breakthroughs. A total of 100 participants from 20 countries attended the conference.

A global overview

AIT's **Dr Krishna R. Salin** highlighted production of the freshwater prawn *Macrobrachium rosenbergii* and the river shrimp *M. nipponense*, a native species of China and farmed primarily there. The major producing countries for *M. rosenbergii* are China, Bangladesh and Thailand. China is the top producer since 1990 and in 2017, contributed almost 50% of global production at 137,300 tonnes valued at USD2 billion.

Salin expressed concern over the production of the giant prawn in Malaysia which stagnated at 250 tonnes since the 1980s. *Macrobrachium nipponense* saw a rapid expansion in production in China, increasing from 87,100 tonnes in 2000 to around 240,000 tonnes in 2017. There is optimism in the future expansion of freshwater prawn farming, fuelled by the increasing consumer demand. "Novel biotechnologies would potentially drive the future of this industry, helping to achieve similar yield levels as those in the marine shrimp sector," said Salin.

Dr Xilin Dai, SHOU in his review on "Research and development work and current status of *Macrobrachium* farming in China" said that *M. rosenbergii* culture was first introduced to China from Japan. Since then, wild populations have been repeatedly imported from Malaysia, Thailand, Myanmar and other countries for breeding and larviculture. Chinese scientists have also developed an improved strain, the South Tailake No. 2.

In 2018, Jiangsu province produced 64,238 tonnes of *M. rosenbergii*. The maximum yield was 9 tonnes/ha. The Yangtze River delta zone produced 88,619 tonnes which is equivalent to 66.5% of the total production, followed by the Zhujiang River delta zone with 37,115 tonnes, contributing to 27.9% while other areas produced 7,532 tonnes or 5.6% of total production.

Broodstocks used are either from the imported wild population, the improved variety South Tailake No. 2 or the all-male and all-female broodstocks from an Israeli company. The number of juveniles produced for grow-out annually is an estimated 30 billion. Larvae are raised in brackishwater constituted from artificial seawater and are fed *Artemia* supplemented with steamed egg and compound feed. Grow-out is carried out using one of three methods: traditional pond culture; polyculture (with vannamei shrimp or fish such as the silver carp or bighead carp) or rice-shrimp culture.

The challenges faced in the giant prawn culture industry include:

- retarded growth which was first detected in 2010;
- lack of high-quality broodstock;

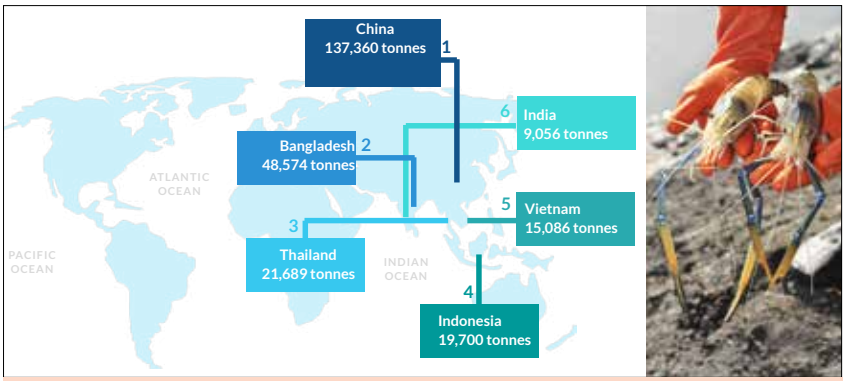


Figure 1. *Top producers of Macrobrachium rosenbergii in 2017 (FAO, 2019) as presented by Krishna R. Salin, AIT, in his keynote on "Freshwater prawn industry – a global overview."*

From left, Guo Qi, Zhejiang Academy of Agricultural Sciences, Hangzhou, China; Weifeng Shen, Zhejiang Academy of Agricultural Sciences; Krishna R. Salin, Asian Institute of Technology, Thailand; Baolong Niu, Zhejiang Academy of Agricultural Sciences; Nyan Taw, Myanmar.

- the need to improve treatment for wastewater and efficient culture methods to improve shrimp quality; and
- more efficient protocols for disease prevention and control.

Research carried out showed that retarded growth in the giant prawn is associated with high water temperatures and increased stocking density leading to an increased proportion of slow-growing shrimp. No significant differences in water chemistry in the culture ponds between normal and slow-growing shrimp were detected. Also, there were no significant differences in genetic parameters including the hybridisation coefficient.

Monosex culture into the genomic era

Dr Amir Segi, Ben-Gurion University of the Nagev, Israel explained that monosex in crustacean culture is desirable due to the differences in growth rate, alimentary needs and behavioural patterns between male and female of the same species. In decapods, the insulin-like androgenic gland (IAG) hormone functions as a hormonal switch; its expression enables male development, and when not expressed female development.

Segi said "For the all-female population, a fully functional sex reversal of females into 'neomales' is required. These 'neomales' are then crossed with normal females to yield progeny containing 25% WW females. Finally, the WW females are crossed with normal males (ZZ) to yield an all WZ genotype female population." On the potential implications of the genomic era on the future of prawn aquaculture and research, Segi said that future genomic research on *M. rosenbergii* will include: genome editing; novel in silico approaches for genes and genomes; identification of gene regulatory regions; genes which do not encode proteins; and epigenetics. Genomic-based selective breeding targeting body size, behaviour, disease resistance, colour and claw, body and head ratios can also be carried out.

Assaf Shechter, Enzootic Singapore shared his company's pioneering technology-driven strategies which he believed can transform the giant prawn industry and its economics. "Commercially available all-female populations are currently enabling a transformational shift to higher stocking densities, higher yields and improved uniformity, thus replacing the traditional focus on the large-sized but territorial males", said Shechter, adding that the high stocking densities of all-female prawns can now support the establishment of pioneering indoor intensive freshwater recirculation aquaculture system (RAS) which can facilitate prawn grow-out anywhere in the world.

On new developments in global farming of all-male *M. rosenbergii*", **Haim Avioz**, Tiran Shipping, Israel said, "The culture of all-male prawns will result in doubling yield and bigger prawns which are the preferred size for the Asian market; such cultures yield more meat for the same amount of time, energy and money invested". The company has two aquaculture farms and hatcheries, one each in China and Vietnam. The technology to produce all-male population uses a cutting-edge approach known as temporal gene-slicing through RNAi which can produce the all-male population. The all-male post larvae are presently sold in China at RMB 1,000-1,200/10,000 post larvae (PL) or USD 142-170/10,000 PL. In 2019, the hatchery in Vietnam sold post larvae to farms in Cambodia, China, Malaysia and Singapore. Sale of post larvae to farms in Asia is expected to reach 2 billion in 2019.

Genetic improvement of the giant prawn

Guoliang Yang, Huzhou University, China in his presentation "Prospects and successes in genetic improvement of giant prawns in China" stressed that the main reason for China's success is the improved giant prawn varieties used as well as the constant innovation and industry development. He said "Presently there are more than 100 hatcheries in China and the annual post larvae production can reach an estimated 25 billion. The annual production in Zhejiang province can be as high as 15 billion, making it the largest post larvae production province in China."

Work on identifying the pathogen responsible for the white tail disease indicated a nodavirus which can be transmitted horizontally and vertically. The South Tailake No. 1 strain, developed in 2014 is specific pathogen free, grows fast and has a high fecundity rate. Tailake No. 2 strain was developed in 2010 via the crossing of the Myanmar and Bangladesh wild populations with the Zhejiang and Guangxi farmed populations. This strain is selected for growth, meat yield, fecundity and disease resistance by using the aquatic animal multi-trait breeding techniques. With constant research and innovation, the Shufeng strain was developed in 2016. Presently, a SPF germplasm bank is being constructed. The excellent growth in the giant prawn culture industry in China is due largely to its three-pronged approach: research to further innovate and introduce new high-quality strains; improvement in the quality of technology transfer and constantly addressing market demand.

Suppatra Uraiwan, formerly from the Aquatic Animal Genetics Research and Development Division, Department of Fisheries, Thailand, spoke on the selective breeding programme on

The Panel on 'What the future holds', from left, Assaf Schechter; Suppatra Uraiwan; Amir Sagi, Donghuo Jiang and Gaofeng Qiu.

The panel on 'Overcoming the regional challenges', from left, Lifat Rahi; Gaofeng Yi, Setya Nandlal (Chair); Nyan Taw; Jeane Rimba Indy.

improving growth rate in Thailand which was first initiated in 1996. The study concentrated on the selection for rapid growth and for different culture conditions. In 1996-2000, a within-family selection procedure was used for growth rate selection. In 2004-2010, tests on growth performance were conducted in the second and fourth generations. Since 2006, the focus is to achieve SPF fry without nodavirus and extra small viruses. Presently, the within family selection has reached the seventh generation and the selected strain is registered as Macro 1 strain.

Technology innovations

Gaofeng Yi, Dabeinong Fantastic Aquaculture Group, China, highlighted the use of technology and innovation by his company in giant prawn culture. He gave three reasons for the occurrence of "iron shrimp" where shrimp growth is retarded and where maturity is attained when the shrimp are at 5-6 cm length. In 'iron shrimp' the claws continue to grow while the rest of the body remain stunted. According to Yi, the first reason was at the nursery stage, selecting female shrimp with more but smaller-sized eggs may eventually cause growth retardation in the offspring. The use of antibiotics at the nursery stage may also have adverse impacts on shrimp growth. On the second reason, Yi said, "When normal shrimp are cultured with 'iron shrimp', the former may also grow slower. This could be due to bacteria and virus in the 'iron shrimp' infecting the normal shrimp. The third reason is related to geography or climate which can play a role in stunting shrimp growth." He noted that 'iron shrimp' is more frequently encountered in the eastern part rather than the southern part of China. "When post larvae are supplied from the same source to a nursery in Huzhou, Zhejiang province, and cultured in two different localities, 'iron shrimp were encountered in greater numbers (60%) in Jiangsu but only 30% in Guangdong provinces."

Health management

Hooper Chantelle, CEFAS, UK discussed work done to determine the pathogens of larval mortality in the giant prawn hatcheries in Bangladesh since 2010. In 2018, larval samples with low survival rates as well as those with mass mortalities were obtained. "The samples were screened by PCR for a range of pathogens known to cause diseases in the giant freshwater prawn. All screened samples were negative for WSSV, MrNV, XSV and MrTV," she said. The most affected samples were used for total shotgun sequencing and a novel 29kb virus was found. Phylogenetic analysis showed that this virus is related to, but very distinct from yellow head virus. This virus is likely to have contributed at least in part to the major decline in post larvae production. Intratubular cytoplasmic inclusion was found in the hepatopancreas of wild giant prawn. Sequencing revealed the inclusions were *Spiroplasma eriocheiris* with a difference in pathogen localisation to *S. eriocheiris* in prawns from Thailand.

Country reviews

Four country review papers were presented. **Dr Lifat Rahi**, Khulna University, Bangladesh stressed the importance of the giant prawn industry in his country. It is the second largest export earning sector which contributes to 3.5% of the country's GDP. But giant prawn production appears to have stagnated. Post larvae production in 2009 was 190 million and since 2013 has remained at 30 million. In the last six years, overall production declined drastically due to the failure in hatchery production throughout the country. Presently, only 3-4 hatcheries out of 46 can produce post larvae. According to **Dr Nyan Taw,** presently, 21 hatcheries in Myanmar, mainly near the Yangon region, produce between 200-400 million post larvae per year. In larviculture, nodavirus and the white tail disease are the two major problems. Grow-out in ponds are usually monoculture or polyculture with fish such as rohu *Labeo rohita* and the common carp *Cyprinus carpio*.

Jeane Rimba Indy, Mexico presented on hatchery production methods and broodstock development for *Macrobrachium carcinus* locally known as la pigua in Mexico. The larviculture of this species, was first carried out in 2009 and was successful in 2015. A multidisciplinary team from several universities from Indonesia, Brazil, India and Israel had implemented a restocking programme for the post larvae produced in the laboratory. The first grow-out

trial rearing juveniles in ponds was carried out in November 2016. **Satya Nandlal**, Queensland University of Technology Australia clarified that live *M. rosenbergii* cannot be imported into Australia since the country has very strict regulations on the import of exotic species. However, the endemic species *Macrobrachium spinipes* found in northern Australia holds great potential for commercial culture. "Until now, development has been constrained by difficulties in closing the life cycle for mass production and there is a general lack of enthusiasm for research into freshwater species in Australia."

What the future holds for *Macrobrachium* spp in an era of biotechnological innovation?

Panel members were in agreement that the quality of the broodstock is of the utmost importance. The focus should be on how to maintain the genetic diversity, improve on the strain for culture and overcome the problem of slow growth syndrome. Genomics and gene editing present great promise. Some participants felt that there is a lack of cooperation among Asian countries in sharing of broodstocks. Generally, there is poor broodstock management or no management at all by individual farmers. Should broodstock management be done by the government agencies in individual countries and then post larvae disseminated to farmers as is done in Thailand and China?

On disease management, especially for the emerging ones, farmers are often unable to identify diseases and successfully mitigate them. There is a general lack of training by field staff to help farmers. The panel suggested that government agencies should give more information to farmers on how to mitigate disease and other farming problems. The strategies adopted by China and Thailand linking government agencies to farmers provide best farming practices to the industry. This strategy is unfortunately lacking in many other countries.

Overcoming the regional challenges that limit the expansion of freshwater prawn farming

New biotechnology and genomics breakthroughs such as all-male and all-female populations will give a boost to this industry. China appears to lead the world in this aspect. The government policy in China "centred on people development for the people by the people" had contributed to the success of its aquaculture industry, which is more profitable than planting wheat or rice. Well-trained staff and development of technology and science innovation are critical to aquaculture development; in China 65% of universities have an aquaculture major. The panel felt that AIT should provide more training courses to farmers, including poor farmers.

It was suggested that for future meetings, government officers, FAO officials and researchers from the WorldFish Center should be invited to attend. Recommendations from this conference should be disseminated to relevant government departments of the various countries and international agencies.

R&D in various fields such as to further understand disease problems like the cross-linking effects of pathogens; protein contents in feed viewed in terms of eutrophication; and broader scale selective breeding should be prioritised. Genomic research was regarded as the game changer which will propel the industry into greater heights.

The next Giant Prawn 2022 will be held in Bangkok from 14–17 March 2022

From left, Quanxin Gao, Huzhou University, China; M. Thanabal, Eck Hatchery, Malaysia; Valencia Yi, New Horizon Aquaculture Pte. Ltd, Singapore; Jian Jin Khoo, STAC, Malaysia; Boo Lai Liaw, KGC Eco Resort Sdn. Bhd, Malaysia; Eng Wah Khoo, STAC, Malaysia; Qiongying Tang, Huzhou University, China; Victor Newn, HHMS Farm, Brunei.

Dr Lifat Rahi, Bangladesh stressed the importance of the giant prawn industry in his country.

Krishna R. Salin said that novel biotechnologies would potentially drive the future of this industry.

Study proves that ICC Brazil products are EHP-free

ImmunoWall® and StarYeast® produced by **ICC Brazil** were tested according to international evaluation standards and obtained negative results for *Enterocytozoon hepatopenaei* (EHP), a microsporidian parasite that affects the hepatopancreas of shrimp. The microsporidian is not highly deadly, but it does impact the growth and performance of animals. Its history of occurrences is mainly in Southeast Asia. One of the means of contamination of these animals is orally, mainly through feeding, this means that if the food is contaminated, the animals will certainly have problems.

Recent research showed that more than half of the raw materials of marine origin for shrimp feed had been tested and the result has been positive for EHP in many countries. ICC Brazil reinforces the need for having safe options to avoid damage to the health of the animals, to the producer or the final consumer.

"Our customers' concerns are our main priority, which is why we submitted samples from ImmunoWall® and StarYeast® to a renowned laboratory in Thailand to detect EHP. The results showed that EHP was "undetectable" for both products through nested-PCR, which means that they are safe options from a health standpoint, says Dr Melina Bonato, R&D Manager.

ImmunoWall® is a natural source of MOS and β-glucans, derived from a thick cell wall of purified yeast. It improves intestinal integrity and promotes microbiota balance due to its efficiency in agglutinating pathogenic bacteria. Its components enhance the immune system. StarYeast® is an inactive dried yeast derived from the fermentation of sugar cane molasses. In addition to high palatability, it has an excellent nutritional profile, with a high concentration of B vitamins.

ICC Brazil is a company that creates solutions for animal health and nutrition. Recognised in Brazil and abroad for its innovative capacity, the company creates products based on sugarcane yeast. Since 1992, ICC has been working with universities and national and international research centres, and over 200 studies and in vivo research have been carried out in different areas of animal production, ensuring the quality and efficiency of our products. www.iccbrazil.com

Xelect Express for rapid response range of genetic services

Since 2012, **Xelect** have been trusted by major producers globally to manage their breeding programmes and provide critical support services. In response to the high demand from the industry, the company has announced the launch of Xelect Express – the new rapid response range of genetic services. Xelect Express offers quick, cost effective access to genetics expertise for genotyping, pedigree assignment, gene expression, ploidy and sex determination.

CEO Ian Johnston commented "Our breeding programme customers have always had access to our genetic testing services, but recently we've seen demand increase. Whether it's impartially checking the ploidy of eggs or conducting spot checks to avoid in-breeding in broodstock, people want to have certainty over the quality of their supplies and the long-term health of their stocks".

The tests will be available globally, and conducted by Xelect's highly qualified experts, led by Operations Director Tom Ashton, who said, "Our teams use the latest technology and insights to offer fast turnaround, guaranteed technical excellence, total confidentiality and – of course – a low price".

In detail, the services offered are:

- Ploidy determination: This 24-hour service is valuable for fish breeders looking to improve their processes or to certify their product and for farms looking to check the quality of their supplies. Xelect can issue a next day Certificate of Ploidy.
- Sex determination in salmonids: An excellent way to manage stock control which allows sex to be determined at the earliest stages. Results usually returned within 10 days of sample receipt.
- Genotyping and relatedness analysis: The genetic analysis of broodstock enables problems associated with inbreeding to be avoided and ensures the long-term sustainability of a breeding programme.
- Gene expression: Xelect offers gene expression measurements to help with the development of new feeds and vaccines.

A wide range of high-resolution images are available on demand. Xelect is the leading independent provider of genetic services to aquaculture and has customers worldwide. It operates a private, fully equipped genetics laboratory in St Andrews, Scotland. www.xelect.co.uk

Expansion and upgrade of world-renowned innovation and development centre

Wenger Manufacturing, Inc., has begun work on a USD13 million renovation project to expand and modernise the Wenger Technical Centre in Sabetha, Kansas, USA. The centre will remain operational through the year-long renovation, thanks to innovative building design and construction methods.

The Wenger Technical Centre is a facility dedicated to innovation and continuous improvement of extrusion process systems for food, feed and industrial applications. It houses full-scale extruders, dryers and ancillary components to provide a real-world development environment for extrusion-based products and processes. A recognised proving ground for innovation and training, the centre is used by clients, academia and other industry partners for accelerating product development and operational training.

"This renovation is a strategic reinvestment into a facility that has long served our industries as the critical hub for innovation and continuous improvement," said Lafe Bailey, Co-CEO and President of Sales and Corporate Development. "The technical centre is of global importance to the extrusion industry since 1954, and we are committed to both renewing, and expanding, the roles and responsibilities that the Wenger Technical Center holds in the industries we serve."

The new construction will increase the existing 22,000 ft^2 (2,043m^2) capacity by 40%, making the facility more scalable and extending its lifespan long into the future. The modernisation will include enhancing preventative measures for food safety. The added area will make the center more versatile. It will expand the scope of market-facing services while also further enhancing existing innovation projects already active in the Wenger pipeline.

"As our industries face increased scrutiny over food safety, the new Centre will provide a low-risk environment to evaluate prototypes and make sure new products and processes adhere to safety and quality standards," said Brend King, Vice President and Technical Center Director. "We're very excited for this expansion as it will allow us to continue helping clients—and Wenger—move ideas from concept to market more quickly."

By operating commercial-scale equipment, Wenger has the ability to scale up production runs to achieve real-life capacities, which is usually not possible in an R&D environment. "By staging a full-scale production setup, clients can truly see what these machines can do, and they don't have to settle for a projection of performance based on calculations," said Bailey. "No other innovation centre of its kind has this breadth of capabilities."

The approach to this renovation is unique in that the new building will be constructed over the top of the existing facility while operations continue inside. The first phase is construction of a modern, tension fabric structure that will completely enclose the original building. The second phase entails taking down the original steel structure underneath to create a new space with greater height and depth as well as a greatly improved process flow. The larger capacity will fit full-scale, commercial versions of nearly every piece of equipment Wenger manufactures, whereas today it houses six extruders and two dryers.

"This innovative approach originated from our need to build a world-class innovation and development facility, all while maintaining continuity of current customer and Wenger projects," said King. "That continuity is important because the market relies on this facility to meet their goals, and we are committed to keeping it open throughout this process."

King added that the company believes this is a responsible approach that clearly aligns with the Wenger culture of "Integrity, Ingenuity and Initiative". "Instead of tearing down valuable infrastructure and starting over at another site, we're repurposing a well-functioning and widely utilised facility that's already positioned right where we want it."

"Wenger also is very grateful to the City of Sabetha and the surrounding community, both of which have been exceptional partners since Wenger was first established here in 1935," said Bailey. Construction began in November and normal operations will run in parallel with construction until project completion during the fourth quarter of 2020.

Wenger is the world's leading supplier of extrusion cooking systems for food processing with a rich 85-year history. With nearly 500 employees worldwide, Wenger operates multiple plants, a world-class innovation and development centres and sales and service offices around the globe. Privately held and headquartered in Sabetha, Kansas, Wenger also has operations in Galena, Kansas, Belgium, Denmark, Taiwan, Brazil and China. www.wenger.com

Virtual experience for 2020 ONE: The Alltech Ideas Conference

Alltech has been closely monitoring the COVID-19 outbreak, with consideration for ONE: The Alltech Ideas Conference (ONE). The annual event was scheduled for 17 -19 May in Lexington, Kentucky, and typically assembles more than 3,500 attendees from 70 countries for an exploration of innovative solutions across the global food supply chain.

Considering rising health concerns related to coronavirus, Alltech will present ONE session topics online, transitioning to a virtual experience instead of a live event in 2020. "Our first priority remains the health and safety of attendees, our colleagues and the communities in which we live and work," said Dr Mark Lyons, president and CEO of Alltech. "With that in mind, we have decided to host this year's international conference on a virtual platform, allowing registrants from around the world to engage in industry-leading content in a way that is accessible for everyone."

The Alltech ONE Virtual Experience will provide access to agricultural topics, including agri-business, aquaculture, beef, crop science, dairy, the future of food, pig and poultry. Live-streamed keynote presentations and on-demand video content from some of the world's leading industry experts, including the most impactful presentations from past years' events, will be available beginning 18 May, 2020. https://one.alltech.com/

New R&D centre for aquaculture in Singapore

ASA (Aquaculture Station by Adisseo) is Adisseo's first research and development dedicated to aquaculture. It will focus on nutrition, aquatic animal health and innovative aquatic science technologies. The grand opening of ASA located at the Singapore Food Agency (SFA)'s Marine Aquaculture Centre (MAC) on St John's Island was held in December 2019. The guest of honour at the opening, Dr Tan Lee Kim, Director General of Food Administration and Deputy CEO, SFA was accompanied by Singapore's Economic Development Board (EDB)'s Director Chemical & Materials, Khalil A Bakar, and Jean-Francois Roux, Executive VP Innovation, Adisseo. Tan's message was to leverage on science and technology to grow more with less. For EDB, aquaculture and urban farming are exciting areas for growth in Singapore.

Ribbon cutting for ASA by Dr Tan Lee Kim, Director General of Food Administration and Deputy CEO, SFA (centre), Khalil A Bakar, Director Chemical & Materials, EDB and Jean-Francois Roux, Executive VP Innovation, Adisseo (left).

At the heart of Adisseo's R&D are seven centres worldwide, each with its own specialisation from analysis, nutrition, formulation, biotechnology, chemistry to chemical process and innovation. Adisseo allocates almost 4% of its annual revenue to R&D and has around 200 people dedicated to research, to continuously bring new and more effective products to market.

During his presentation on ASA, Roux, said, "In the value chain, Adisseo aims to deliver the right products for the right species. This is through research and innovation. Singapore, where there is Adisseo Asia-Pacific headquarters, was chosen for this R&D centre for several reasons. SFA and EDB have been very supportive with advice since day 1 and with strategic investment." He added that Singapore's innovation funds have been actively increasing the aquaculture footprint and fostering the country as a hub for innovation in aquaculture. "In Singapore, we can be assured of IP security and there is a strong focus on sustainable and urban aquaculture. There is also a strong research ecosystem with partners – Temasek and Republic Polytechnics, A*Star (Agency for Science and Technology) and James Cook University."

ASA has 72 round tanks, each of 500L capacity that can be partitioned to reduce the volume to 150-250L/tank to be used for shrimp trials. Photo credit: Adisseo

Dr Stefan Jakob, Director R&D, Nutrition and Animal Health explained how Adisseo approaches innovation by developing platforms and partnerships. "In some areas, it is done in-house such as at CERN in France; others are tailor-made within the needs and proposals from different partners or where Adisseo has researchers working on an on-going project. Researchers can pitch for funding to carry out research. Now we have ASA for aquaculture research."

David Bal said that the automation in water cleaning at the drum filter will avoid water quality problems. Photo credit: Adisseo

With ASA, it has been a long journey for Jakob and David Bal, Head of Research, Aquaculture Nutrition and Health, Adisseo Asia Pacific, in setting up this state-of-the-art research facility in Singapore. Bal explained that to ensure functionality, the actual system designed for ASA was setup first in Germany, tested, dismantled and then was set up in November 2019 in Singapore. "What we have is a flexible system to meet customers' needs, in fish and shrimp aquaculture research. The key for any experiment is statistics.

To ensure this and flexibility, David Bal explained, "We have a total of 72 round tanks, each of 500L capacity and which can be partitioned to use for shrimp trials, whereby we can reduce the volume to 150-250L/tank). All tanks have faeces collectors for digestibility studies. In addition, we have a quarantine system to prepare test animals. We can use as many tanks as we need and

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Dr Stefan Jakob (left) and David Bal (right) describing the tank setup at ASA for aquaculture research during the visit to facilities after the opening ceremony. Photo credit: Adisseo

adjust temperature, salinity and control flow rates in each unit. Having many tanks allow us to conduct studies with a high number of replicates and treatments and obtain quality data."

There are options to select freshwater or marine recirculation systems depending on requirements. The tilapia is a future target species. For water quality monitoring, Bal added, "We use an automated water quality monitoring system and will develop more use of technologies such as IoT and AI (internet of things and artificial intelligence). These technologies reduce the use of manual labour as well as human errors in water quality management. The whole system takes into consideration the need to minimise labour and conserve energy and reduce costs as both are costly in Singapore."

Adisseo is one of the world's leading experts in feed additives. The group relies on its research centres and its production sites based in Europe, USA and China to design, produce and market nutritional

Teams from Adisseo's Aquaculture, ASA, APAC and R&D at the opening of ASA. Photo credit: Adisseo

solutions for sustainable animal feed production. Adisseo is one of the main subsidiaries of China National BlueStar, leader in the Chinese chemical industry with nearly 21,500 employees and a turnover of USD9.3 billion. Adisseo is listed on the Shanghai Stock Exchange. www.adisseo.com

Calysseo: Joint-Venture to commercialise an innovative feed solution for aquaculture

Bluestar Adisseo Company (**Adisseo**), a world leader in feed additives for animal nutrition and the world's leading alternative protein producer, **Calysta**, have signed an agreement to establish a 50:50 joint venture named **Calysseo** to develop FeedKind®, an innovative feed solution for aquaculture, and to provide exclusive supply for Asian markets.

The strategic partnership will also see the construction in China, of the world's first commercial FeedKind® production facility. The first phase is expected to start operating in 2022, delivering 20,000 tonnes of FeedKind® protein per year. This is a major business supplying aquafeed ingredients in Asia with a market size of USD28 billion, which represents 70% of the global market.

The second phase of the partnership will add a further 80,000 tonnes of capacity to the site, allowing for extended market penetration leading to further development to follow customer demand. Jean-Marc Dublanc, CEO, Adisseo said, "Adisseo is committed to strategic investments in new disruptive technologies. Our ambition is to become one of the leaders in sustainable feed ingredients and additives for aquaculture across the Asian markets. For this reason, we have been investing significantly to develop an aqua lab station in Singapore. With this joint venture, we are investing further in a strategic collaboration, combining the agility of a start-up with Adisseo's expertise to enable Adisseo to become a major contributor to food safety and sustainability in Asia via innovation."

Dr Thomas JG Huot, Chief Operating Officer, Calysta, said, "Adisseo is one of the world's leading feed additive companies and this exciting new partnership with Calysta marks a significant moment in the FeedKind story. This announcement means we are now making the transition from a tremendously exciting idea to one that will deliver truly world-scale impact, making food supply chains more resilient and making them far more sustainable in the process."

Through this strategic partnership, Adisseo is tackling one of the key challenges faced by the aquaculture industry – to provide high quality seafood without adding extra pressures to wild fisheries and the environment. FeedKind® is made using very little water and no agricultural land by fermenting natural gas, an abundant source of energy, to create a safe, nutritious, traceable and affordable protein. FeedKind® is price competitive with existing protein sources and produced to the highest quality standards.

Jean-Marc Dublanc added: "Adisseo looks forward to developing this partnership with Calysta, as this innovative technology will support and accelerate the development of its business in aquaculture while providing additional opportunities to tackle the dual challenge of meeting the world's need for healthier, safer, affordable and sustainable food for tomorrow while at the same time contributing to protect the planet's resources." www.adisseo.com; www.calysta.com

Towards an ultimate sensor of nitrogen compounds in water

Nitrogen Sensing Solutions, NS2, is a Portugal based startup founded by three scientists, Gabriela Almeida, Anjos Macedo and Ana Viana, in August 2019. The company's first product, NOxAqua, is the result of 20 years of research that is now customised for the aquaculture market. NOxAqua is a smart multiparametric test for water analysis, measuring simultaneously, accurately and in real-time, three nitrogen compounds - ammonium, nitrite and nitrate – that can accumulate in either ponds or tanks, and above certain levels, become highly toxic to fish.

"Our technology is completely different from what is currently used in the market. Most methods are not very accurate, like the colorimetric ones, requiring chemical standards and calibration procedures, which can be cumbersome and expensive. There are very few quantitative solutions that provide results in real-time, but are rather expensive and unable to accurately differentiate between nitrate and nitrite compounds. Initially we were totally focused on the salmon farming in RAS (recirculating aquaculture systems), as early adopters, where monitoring such chemical parameters is very critical. But now in Asia, we are aware that there is also a market demand in shrimp farms", said Viana.

Gabriela Almeida, Anjos Macedo (centre) and Ana Viana (left). The company's first product, NOxAqua, is the result of 20 years of research and has been customised for the aquaculture market. It is a smart multiparametric test for water analysis, measuring simultaneously, accurately and in real-time, three nitrogen compounds - ammonium, nitrite and nitrate.

The innovative technology provided by NS2, NOxAqua, consists of an electrochemical biosensor that recognises specifically, the nitrogen compounds, in less than 5 minutes, and transmits a signal proportional to the concentration of each compound in water. "NOxAqua is a highly sensitive hand-held device, a reader, that connects disposable strips to measure nitrogen parameters in a drop of water. Data can be sent to a phone, laptop or to the farm dashboard that usually collects other water quality parameters (dissolved oxygen, salinity, redox potential and pH). We want to make our sensor very straight forward to the farmers. If they use our product, they can get accurate results quickly and access the water quality immediately, at any time.

"Today, we have a prototype developed for nitrite detection, that we are selling to a Norwegian company. Here in Asia, we have also made good contacts for product scale up. We are now seeking investments for the next steps and to innovate further in the measurement of nitrate and ammonium. Our goal is to have a fully automated system to quantitatively measure all three nitrogen compounds, using a triplex sensor," added Macedo.

When NS2 first started through an entrepreneurship programme in Portugal, HiTech, the founders learnt at that time that aquaculture could be an important market. The Hatch Programme validated the strong need for their product in the aquaculture market and they got acquainted with worldwide key players in this field. www.nitrogensensing.com

Appointment

COO to lead global operations

Thomas JG Huot, Chief Operating Officer, Calysta

Leading alternative protein producer **Calysta** has appointed Thomas JG Huot, PhD. as its Chief Operating Officer to lead global operations and corporate development activities. Additionally, Huot is responsible for corporate strategy and the commercialisation of Calysta's FeedKind® protein. Huot brings more than 20 years of experience in research in biological sciences, venture investing, project financing and strategic partnerships, including a significant focus on industrial biotechnology, specialty chemicals and nutrition. Huot joined Calysta in 2018 as Chief Business Officer.

"With FeedKind protein, Calysta is well positioned to address the dual challenge of food security and environmental sustainability. I'm delighted to have joined at a particularly exciting time in the company's trajectory," said Huot. "I am looking forward to expanding Calysta's innovations to broader markets, particularly in Asia where there is significant opportunity to provide feed products for aquaculture." www.calysta.com

Heterotrophic microalgae to produce astaxanthin

Hawaii-based **Kuehnle AgroSystems** or KAS is a biotechnology startup which brings to market specialty chemicals and raw materials (such as astaxanthin) for animal markets (feed, colourant) and for the human market (cosmetics, nutrition). Of interest to aquaculture is the natural astaxanthin produced in dark fermenters. KAS uses its extensive library of Hawaiian microalgae strains and exotic plant cell cultures (plant "stem cells") to provide unique cross-disciplinary expertise in cell line creation, target maximization and process scale-up methodology. This is perhaps the largest privately-held collection of Hawaiian microalgae in the world.

"Industrial fermentation ingredients cultured for highest efficiency and quality enables our partners to gain rapid access to markets. KAS works with its partners and customers to originate new concepts, and to discover and deliver preferred bioactives, colourants and other biofunctionals to their markets. For the aquaculture and aquafeed markets, KAS has NATUREKROME™ which is a low-cost natural colourant for aquafeeds produced from whole cell algae," said Dr Adelheid Kuehnle of KAS. She is co-founder and President, who has more than 30 years in plant science research and is part of the 6-member team of this startup company. Adelheid and Gordon Wallace, CAO, joined Hatch Blue's 2019 mentor programme, travelling from Hawaii, Norway to finally Singapore in November 2019.

"The market for colourants in fish and shrimp is dominated by the expensive synthetic astaxanthin. There are also new producers using outdoor production systems such as in the Atacama desert in Chile and in Australia as well as those in China. There is also the question of what is the percentage of the active ingredient," added Kuehnle.

Adelheid Kuehnle

"We produce whole-cell dried algae *Haematococcus pluvialis* in a patent pending process of dark aerobic dark fermentation at very high density, thus lowering the production costs and raising quality. This process is scalable and deployable around the world. It does not require any extraction, has no waste and no polluting run-offs."

The opportunity for KAS's astaxanthin is in replacing synthetics with natural colourant. This is important for the organic feed industry where synthetics astaxanthin is not permitted. KAS is also targeting feed manufacturers and vertically integrated operations who want to switch to natural ingredients, perhaps to boost their feed brand, follow the demand of retail markets or just be ahead of competitors in the premium feed market. The ingredient delivers 2% astaxanthin, 2.5% total pigment. The product is stable under recommended storage and has nominal loss under conditions seen in extrusion or pelleting.

"Via the Hatch programme, the traction was made with ingredient sales and strategic partnering with some of the leading global feed producers, integrators and seafood marketeers," said Kuehnle.
www.kuehnleagro.com

Appointments

First president transition

Kemin Industries, a global ingredient manufacturer, has appointed Leo Xie-Lei as President of **Kemin AquaScience™**, the company's global business unit that serves the aquaculture industry with sustainable solutions. Leo succeeds KP Philip, the first president of Kemin AquaScience™ who will continue to lead strategic projects for Kemin Industries. Leo joined Kemin AquaScience™ as chief commercial officer in late 2018. He also served as regional director for China, building the Kemin AquaScience™ business there from the ground up.

Leo Xie-Lei President of Kemin AquaScience™

"Leo joined Kemin AquaScience™ when it was just beginning, and he has been an asset and a leader in the business since day one," said Dr Chris Nelson, President and CEO, Kemin.

"Being part of the Kemin AquaScience™ team since its launch has been a tremendous experience, and I look forward to continuing our strong momentum as we expand our product portfolio and global reach," said Leo who is currently based in Shanghai and will continue to lead Kemin AquaScience™ from China.
www.kemin.com

First Chief Scientific Officer with NGS expertise

Dr Han Ming Gan has joined **GeneSEQ Sdn Bhd** as the first Chief Scientific Officer, since March 1, 2020. Gan will bring in nearly a decade of expertise in next-generation sequencing (NGS), microbiology, genomics and computational biology from industry and academia. He has a strong background in Illumina sequencing technology from three years of industry experience, as an application scientist at Science Vision Sdn Bhd and from operating two NGS labs during his post-doctoral tenure at Monash University Malaysia and Deakin University Australia. Gan has experience in sequencing and analysing the genomes of Asian Arowana, Australian Murray Cod and black tiger prawn.

In addition to assembling the first complete genome of a Malaysian *Vibrio parahaemolyticus* strain using Oxford Nanopore long read, Gan has also led and recently published the comparative genomics of 40 *V. parahaemolyticus* associated with shrimp aquaculture in Malaysia.

Han Ming Gan is the first Chief Scientific Officer at GeneSEQ Sdn Bhd

WELCOME

Cork, Ireland, September 29 – October 2

aquaculture europe 20

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For more info on the CONFERENCE:
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The Blue and the Green

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Phasing out South American soya in feeds

Denmark's feed company **Aller Aqua** announced that it is phasing out the use of South American soya and focuses on purchasing from regional markets. It wants customers to have high quality feeds with stable performance, but also with minimal environmental impact." In recent years, there has been a lot of focus on soya produced in South America, and the derived effects thereof, such as deforestation and cultivation methods. In our work to continuously increase sustainability and purchasing raw materials in proximity to our European factories, we will now phase out the use of soya from South America," explains Henrik Halken, Group Vice President, Aller Aqua Group.

"Our four European factories have already begun this process. In 2019, 50% of the soya we used in our European factories were regionally produced, and during 2020, this number will reach 100%. This is completely in line with EU initiatives supporting an increased production of protein crops to increase our self-sufficiency – and thereby reduce import. We work actively with the UN Sustainable Development Goals in all our factories. Raw materials are a significant part of the production process in aquafeeds, and it is therefore natural to focus on reducing environmental impact here." www.aller-aqua.com

First super-intensive indoor shrimp farm in Qatar

Blue Aqua International and ITQAN have announced a partnership to establish the first super-intensive indoor farm in Al Areesh – Shamal in North Qatar, to produce 1,000 tonnes of shrimp annually. The project includes a hatchery, grow-out modules and a microbiology laboratory. It will showcase Blue Aqua's knowhow in shrimp farming – including farming methods using the group's patented Mixotrophic™ System and its latest innovations including Green-to-Clean recirculation aquaculture system and the AgroDome™ housing technology. ITQAN Chairman Musallam Al-Nabit said, "The long due diligence to determine the company and system that will best safeguard our nation's land and water resources was time and effort well spent. My pledge is that we will ensure every shrimp our partnership produces will be the most exquisite the Qatari market will ever enjoy,"

Dr Farshad Shishehchian, CEO/Founder, Blue Aqua International said. "This new partnership will leverage on ITQAN's operational competence and Blue Aqua's technical knowledge to realise the production output and a model farm for the region. We have developed a cost-productive super-intensive culture method and farm system design capable of efficient utilisation of water, energy and space. www.blueaquaint.com

AQUA CULTURE Asia Pacific in 2020

Volume 16 2020				
Number & Month	3 - May/Jun	4 - Jul/Aug	5 - Sep/Oct	6 - Nov/Dec
Aqua Business *Feature articles and contributions from industry players*	Experiences from industry and opinion articles covering role models, benchmarking, health management, SOPs, social investments, CSR, ancillary services, self-regulation etc			
Issue focus *Recent developments/spotlight on emerging challenges*	Hatchery	Sustainable & Responsible Aquaculture	Demand & Supply Equilibrium	Aquaculture Education
Industry Review *Developments, outlook, demand & supply*	Aquafeed Production	Tilapia	Aquaculture Start-ups	Catfish & Freshwater Fish
Feeds & Processing Technology *Technical contributions from industry*	Lipid Nutrition	Health Nutrition	Larval & Nursery Feeds	Processing Technology/ Feed Safety
Production Technology *Technical information along the value chain*	Hatchery Technology	IOT/Innovations	Post-Harvest Technology/ Processing	Organic Aquaculture
Marketing activities	Market and product development, market access, certifications, branding, food safety etc			
NEW Post Harvest Quality & Processing	**Technical contributions from industry players on assuring quality at pond site to processing technology**			
Company/Product news	News on activities at international, regional and local conferences and trade shows			
	Deadlines			
Technical articles	16 Mar	11 May	13 Jul	14 Sep
Advert booking	27 Mar	22 May	24 Jul	25 Sep
Show Issue & Distribution at these events as well as local and regional meetings **Show preview*	**VICTAM Asia and Animal Health & Nutrition Asia 2020** Bangkok, Thailand July 9-11	**TARS 2020: Shrimp Aquaculture** Ho Chi Minh City, Vietnam August 19-20	**Taiwan International Fisheries and Seafood Show (TIFSS 2020)** Kaohsiung, September 24-26	***World Aquaculture 2020 Singapore** December 14-18

WELCOME

Singapore - December 14-18, 2020

Singapore EXPO Convention and Exhibition Centre

NEW DATES

WORLD AQUACULTURE Society

WORLD AQUACULTURE SOCIETY ASIAN PACIFIC CHAPTER

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For more info on the CONFERENCE: www.was.org - worldaqua@was.org
For more info on the TRADESHOW: mario@marevent.com

Announcement on new dates:

3-5 June 2020, Can Tho City, Vietnam

VIETSHRIMP Aquaculture International Fair 2020 (originally scheduled for 25-27 March) has been rescheduled to 3-5 June 2020. The location is the Can Tho Promotion Agency, 108A Le Loi Street, Cai Khe Ward, Ninh Kieu District, Can Tho City, Vietnam. The organising committee will monitor the situation with COVID-19 closely and will provide details of the rescheduling arrangements to all exhibitors and visitors.

For enquiries, the service hotline is (+84) 944663828 (Mr. Nghia), Email: vietshrimp@gmail.com. https://vietshrimp.net/reschedule-notice/

9-11 July 2020, BITEC, Bangkok, Thailand

Heiko M. Stutzinger, VIV worldwide Director and Managing Director VNU Asia Pacific together with Sebas van den Ende, General Manager, VICTAM Corporation announced that **VICTAM and Animal Health and Nutrition Asia 2020** has been postponed to 9 – 11 July 2020. Organiser assessed that the actual situation will not guarantee the quality of the exhibition and will not create enough business opportunities for exhibitors and visitors. Over 80% of the exhibitors agreed with the organisers that the postponement of the event is the best solution and have therefore confirmed the decision to move their participation to July 2020, trusting that the situation will improve during the next few months. www.victamasia.com

Cancellation

Organisers of **Aquafeed Horizons 2020 conference**, the specialised conference for aquaculture feed professionals due to take place March 24, 2020 in Bangkok, have cancelled the meeting.

"This was a very difficult decision, and we waited as long as we could in the hope that the coronavirus epidemic would slow, but unfortunately we are not seeing that", Suzi Dominy, publisher of Aquafeed.com said. "We are sorry to disappoint our delegates but their safety and wellbeing, as well as that of our presenters and staff has to come first. It would have been irresponsible to go ahead with an international meeting in this region; we really didn't have a choice." www.feedconferences.com

2020

Details on the events below are available online at **http://www.aquaasiapac.com/news.php**
To have your event included in this section, email details to **zuridah@aquaasiapac.com**

June 3-5
VietShrimp 2020
Cantho City, Vietnam
www.vietshrimp.net

Postponed
(TBA on new dates)
Seafood Expo Global 2020
Brussels, Belgium
www.seafoodexpo.com

May 28-30
Livestock Philippines 2020
Pasay City, Philippines
www.livestockphilippines.com

July 9-11
VICTAM Asia and Animal Health & Nutrition Asia 2020
Bangkok, Thailand
www.victamasia.com
www.vivhealthandnutrition.nl

August 19-20
TARS 2020: Shrimp Aquaculture
Ho Chi Minh City, Vietnam
www.tarsaquaculture.com

August 26-28
Vietfish 2020
Ho Chi Minh City, Vietnam
www.vietfish.com.vn

August 31-September 2
VIV MEA 2020
Abu Dhabi, U.A.E
www.viv.net

September 8 – 10
Livestock Malaysia 2020
Melaka, Malaysia
www.livestockmalaysia.com

September 24-26
Taiwan International Fisheries and Seafood Show (TIFSS 2020)
Kaohsiung, Taiwan
www.taiwanfishery.com

September 29-October 2
Aquaculture Europe 2020
Cork, Ireland September
https://aquaeas.eu/

September 29-October 2
11th Symposium on Diseases in Asian Aquaculture (DAA11 2020)
Kuching, Malaysia
www.daa11.org

December 14-18
World Aquaculture 2020
Singapore
www.was.org

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