



THE
AQUACULTURE
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Shrimp
& Aquaculture
The New Normal

TARS 2016 Meeting Report

TARS 2016: Shrimp Aquaculture & The New Normal, the sixth in The Aquaculture Roundtable Series was held in Phuket, Thailand from 17-18 August. This was a follow-up to TARS 2014, held at the height of the early mortality syndrome in Asia. Some recovery was expected and in 2014, participants suggested options for the 'recovery, revival and renaissance' of the industry. This has not been realised and the industry remains vulnerable to disease outbreaks and disease threats to the industry have multiplied.

The program for TARS 2016 was structured to take the industry to a new normal with the realisation that disease-free farming is passé. Today, it is all about learning to live with diseases, controlling variables, adapting farm practices and nutrition, and better production planning. Managing diseases should not be left to only the farmers but is the responsibility of all stakeholders.



Juadee Pongmaneerat

In her welcome remarks, **Dr Juadee Pongmaneerat**, Deputy Director General, Department of Fisheries, Thailand emphasised on the need to "work around the diseases" and factor in counter measures in production planning. "The reality is that we need to adapt and change so that Asia's shrimp industry can continue to grow.

"In 2015, Thailand produced 250,000 tonnes of shrimp, which was better than in 2014 but still below our production of 540,000 tonnes in 2012. Our best was in 2010 at 640,000 tonnes. Our leaders in the shrimp farming industry are leading the charge and we are optimistic that soon we will recover."

The State of the Shrimp Aquaculture Industry in Asia - 'Change, Change, and more Change.'



Robins McIntosh

As per its tradition, TARS 2016 started with the state of the industry address. **Robins McIntosh**, Vice President of Charoen Pokphand Foods, Thailand gave an overview on recent developments and what is required for future sustainability.

"Historically the industry has had four epochs and two crises," said McIntosh. "The good years in 2010 and 2009 were followed by a 20% decline in production in 2013. These are what I call catastrophic losses. We have not seen any improvement in shrimp production for 3 years. In 2016, we can expect 11% less shrimp," he continued.

The history of global shrimp production was typified by the trend in Thailand. "Our first crisis was solved with domestication and biosecurity in 2001. The Golden Age of Shrimp started in 2003, with almost 300% increase in world production over a 10-year period. The second crisis came in 2011 with a levelling of production. Just as we had ended the first crisis with change, so again we are looking at change to end this crisis. I believe that when we solve this, the industry will go into another Golden Age. Change is hard and shrimp farmers tend to not like change, but sometimes change is necessary. Once you find the right change, things get better."

EMS is complicated

"It is not AHPND alone or EHP alone or only WSSV. It could be AHPND with EHP, AHPND and WSSV or AHPND with the bacteria *Shewanella*. All of these pathogens are out there at the same time, and interacting in ways we did not anticipate."

The bacteria *Shewanella* is also present with the same prevalence as AHPND. Together there is a synergistic effect and there is higher mortality than with *Vibrio parahaemolyticus* (Vp) alone as described by Dr Kallaya Sritunyalucksana (see p18).

"With WSSV-AHPND interaction, when well controlled, there can be no mortality when the Vp count is at 10^2 CFU/mL. However, what will the farmer diagnose this as? Usually, this is taken as a WSSV loss. In many countries, we are seeing reports of new WSSV outbreaks, generally not seen 5 years ago."

Post larvae efficiency index

A good gauge of recovery, according to McIntosh is the 'post larvae efficiency index.' This is the amount of post larvae used to produce 1 tonne of shrimp. In 2010, the industry in Thailand used almost 10 tonnes/million post larvae (PL). During the depth of the crisis, this decreased to 3 tonnes/million PL. In 2016, there will be a technical improvement and a signal for recovery with an estimate of 7 tonnes/million PL.

"This year we can produce 320,000 tonnes and by 2017, continue up to 350-360,000 tonnes. I have confidence in this because we understand now that two toxins create AHPND and we know what we are doing now."

The model for recovery from AHPND

"Basically, the bacteria worked with quorum sensing which needs a threshold amount of bacteria. By reducing the amount of bacteria in the pond, farms could reduce the severity of the outbreak. At the same time, I had found that there was a genetic component, a heredity to tolerance. In 2016 in Thailand, we found that if we increase shrimp tolerance, we had better results. Today, as we continue to increase the tolerance of the shrimp to the toxin, we can have production back to the level before AHPND came."

"As we now understand the enemy (AHPND), we can create a new biosecurity rule. This is not to exclude the bacteria, but just to reduce the levels through elimination and reduction of the food and the substrates that make those bacteria grow. These include all shrimp feeds, moulds, sludge and organic material in the pond. In 2012, we showed that if the levels were not above 10^4 CFU/mL, there was no mortality, provided there is only one factor. Exclusion is not necessary but just limit the levels.

In Thailand, there are fewer culture ponds today. In 2010, one farm had 78% of pond area for shrimp culture but in 2016, it has only 38%. The major increase is with reservoir ponds, from 13% to 46%. With these changes, profits are higher, shrimp sizes are bigger and productivity better. Crop failures from AHPND is less.

The big debate on definition of SPF/SPR

Specific pathogen free (SPF) stock refers to the health status of a stock and not a genetic characteristic. To qualify as SPF, a shrimp must be free of all known shrimp viruses. Specific pathogen resistant (SPR) refers to a genetic characteristic, that is being resistant to a specific pathogen; a shrimp may be both SPF and SPR.

"There is a lot of misunderstanding on SPF/SPR such that it has created problems in the industry. SPF just means clean shrimp. This is essential for any biosecurity program and to reduce the risks of translocating shrimp diseases. If we had the discipline with surveillance, EHP would not move around as it has. We also want tolerance to a specific disease, but we should have this in a clean body."

McIntosh also addressed another aspect, that selection for resistance sacrifices growth. "Selection for both characteristics takes a lot more work for families and costs more. However, if I had to choose, I would give up some resistance for growth, because growth is the economical driver here."

"The new normal," said McIntosh is "changing with science and technology, with transparency, and with pride. A profitable shrimp culture for the industry and affordable shrimp for the everyday consumer."

Lessons Learnt Post IMNV in Indonesia

In his presentation on "Indonesian Shrimp Farming, Lessons learnt from IMNV," **Anwar Hasan**, Regional Technical Manager - Aquaculture, Biomin said that when dealing with infectious myonecrosis virus (IMNV), the focus was on maintaining a sustainable carrying capacity, improving culture conditions and reducing stocking density to mitigate the disease.



Anwar Hasan

"With the vannamei shrimp, we changed our culture density, from low density to high and super intensive systems. The IMNV outbreak affected our shrimp farming from 2008-2012 with shrimp mortality ranging from 20% to 80%. In the initial years, the effect was necrosis of the skeletal muscle at 90 days of culture (DOC 90) but gradually, the impact of the disease was 40% mortality with initial symptoms appearing at DOC 40.

The industry in Indonesia attributed the severity of IMNV to carrying capacity and began to work on adjusting culture practices to match pond biomass. In addition, attention was given to reducing water exchange and improving water quality within the pond with probiotics. Biosecurity measures and partial harvesting were introduced. Sludge removal, already a common practice in the early years of shrimp farming in Indonesia, intensified using newer models to increase efficiency and automation. Anwar gave some examples of changes at the farm level.

"In 2007, the stocking density at one farm was 120-150 PL/m². Although, IMNV caused some mortality, levels were low. In 2008, as mortality increased, the farm reduced stocking density to 60-80 PL/m². As mortality continued, stocking density was further reduced to 40 PL/m² in some farms. Shrimp survived with no IMNV. The farm increased stocking density to 50 PL/m² but still IMNV outbreaks did not occur.

"Although, IMNV is no longer a threat today, Indonesian shrimp farmers continue with these 'living with IMNV' farm practices. In hatcheries, the emphasis is on IMNV prevention using disease-free broodstock and post larvae as well as better quality post larvae. At the farm level, treatment of incoming water and holding water in reservoir ponds are carried out. Aeration increased with one HP paddlewheel for every 200-400 kg of biomass in the ponds instead of the previous practice of one HP/500-700 kg of biomass," said Anwar.

"Since 2012, Indonesia's shrimp production has been increasing, fueled by high prices. Unfortunately, farms tend to go back to increasing stocking density and now we have IMNV together with white faeces disease (WFD) outbreaks," added Anwar.

The Tsunami of Vannamei Shrimp in India: Challenges to Future Growth



Ravi Kumar Yellanki

Ravi Kumar Yellanki, Managing Director, Vaisakhi Bio-Resources, called the phenomenal growth in India's shrimp farming industry from 2010 onwards as a tsunami. While India's shrimp production grew to 400,000 tonnes within 5 years, production in 2015 dropped 10% to 363,450 tonnes.

"We expect a recovery in 2016 at 400,000 tonnes. This does not mean that we are successful as the increase in production is from new ponds and farms moving to more intensive farming of the vannamei shrimp. In fact, we have more farming areas but productivity has not increased. Our future growth depends on how we address weak links in the supply chain," said Ravi.

"Upstream we have the central quarantine system, screening every batch of broodstock for OIE listed diseases and we have a broodstock multiplication centre (BMC). Small farms (<2 ha) dominate making it difficult for adequate biosecurity measures, although these are easier to operate."

Adequate imported broodstock is critical for the whole production process. This is also to prevent the use of pond-raised broodstock. India, similar to other countries, imports broodstock from suppliers in the US for maturation in hatcheries or post larvae for the BMC. The situation is different in India. Any delays in delivery, means waiting for available space. On arrival, the mortality rate of broodstock can be up to 60%. These and custom duties increase operation costs of hatcheries.

To overcome these problems and meet demand in India and other parts of Asia, Ravi said, "The authorities will set up an additional broodstock quarantine facility north of Chennai and will build three BMCs. There is an opportunity for a breeding company to set facilities in the secluded Andamans," said Ravi.

Larger farms have added a nursery phase in their production cycle but this remains a constraint for small farms. "It is important that hatcheries cooperate with small farms to supply larger post larvae and also post larvae acclimated to low saline pond culture. Three phase farming allows for staggered harvesting. In turn, this will put less pressure on processors," said Ravi.

WSSV, *Enterocytozoon hepatopenaei* (EHP), white faeces disease (WFD) and running mortality syndrome (RMS) are the major disease threats. High and unprecedented losses are from WSSV because of poor biosecurity in small farms. The threat level of WFD is higher than that for EHP, where the prevalence can be 3 out of 20 ponds in a farm. Partial harvesting in infected farms reduces losses. RMS is often reported in Indian shrimp farms.

The final stage in the value chain is marketing shrimp. "When we produce more, where are we going to sell? We need to look for more markets. Our processors need to consider moving into the value added segment. India lacks a domestic market for its shrimp," added Ravi.

Sustainable Shrimp Farming in Latin America with Performance Feeds

According to **Dr Camilo Pohlenz**, Manager, Feed Trial Unit and Product Developer, BioMar Americas, Costa Rica, FAO reported an overall 95% rise in shrimp production in the past ten years in Latin America. The current strategy is to move toward a lower



Camilo Pohlenz

dependence on wild caught shrimp and the new normal is to increase farmed shrimp production. Major producers are Ecuador, Mexico, Brazil and Honduras.

“In 2015, Ecuador led with an industry estimate of 330,000 tonnes. In Mexico, the preliminary estimate by the Ministry of Agriculture was 130,000 tonnes for 2015. For Mexico, this was a significant improvement over the 87,000 tonnes produced in 2014. Overall, a 5% increase in production is expected in 2017,” said Camilo. “In general, culture systems have remained extensive to semi-intensive in large earthen ponds, some as large as 100 ha. Stocking density range from 6-10 PL/m² for extensive to 15-30 PL/m² for semi-intensive culture. Our largest shrimp are 25 g and smallest are 12-16 g. Contrary to the demand in Asia for fast growth, in Latin America, the genetic pool has been driven towards disease resistance rather than growth.

“In the drive for a sustainable industry in Latin America, stakeholders realise that they need a back-up plan when the world wide industry recovers and perhaps also consider a genetic mix of disease resistance and growth together with bio secure, controlled shrimp farming models,” added Camilo.

While disease is a threat to an industry, which does not understand change, Camilo also sees it as an opportunity for the industry to grow and introduce new technologies, production systems and performance-driven feeds. This is the game changer.

“WSSV came in 2002 and we learnt our lesson by lowering stocking density, strengthening sanitary surveillances and changing types of feed. We also became interested in more controlled production systems. In Mexico, before we could solve WSSV, we had outbreaks presumed as early mortality syndrome. The industry implemented alternative farming systems such as easier to manage smaller (1.5 -2 ha) ponds, feeding trays, super intensive culture at 100-150 PL/m² and in Ecuador, recirculation systems. Ecuador picked up production while Mexico lagged behind. With health issues in Mexico in 2013, some farms changed to include three phase controlled production, obtaining very promising results,” said Camilo.

According to Camilo, Latin American shrimp farmers still demand fish meal in shrimp feeds as well as familiar raw materials. The feed producer has to see how this fits into the future need for sustainable feeds. At the same time, he sees disease as an opportunity for performance-driven feeds.

In his presentation, he showed data on nutrient and essential amino acid availability in marine and vegetable meals. He also showed some trials on biorhythmic feeding of shrimp and made growth comparisons with crude versus available protein in diets. “Still there are important ‘unknowns’ in shrimp nutrition and different players may dictate the ‘degree’ of a given nutrient requirement based on size of shrimp, water conditions, density and special conditions such as the disease state. This will remove the reliance on ‘historic’ feed ingredients. Special attention must be put under ‘high pathogenic pressure’ and use of specific additives could ameliorate the negative effects of diseases.”

The Lost Billions in Asia’s Shrimp Aquaculture Industry

Andy Shinn, Senior Scientist, Fish Vet Group Asia Ltd, Thailand described ways to calculate losses. In Thailand, a simple way is

using the average price of shrimp; the loss was USD 5 billion since 2011 which may be attributed to AHPND.



Andy Shinn

Using average prices/kg/day, we have USD 7.4 billion being lost. We cannot attribute all losses to one pathogen as we know that there are co-infections. When we translate these into real shrimp terms, we have an astonishing 3.5 trillion shrimp. In Thailand we see that productivity fell from 10.6 tonnes/ha in 2010 to only around 4.13 tonnes/ha in 2013 with AHPND outbreaks.”

In Vietnam’s Mekong Delta, to estimate losses, he used production areas with some assumptions on culture system intensity, area and stocking density. “In the case of AHPND, the assumption was 50% mortality of shrimp within 20 days of culture. Calculated losses from AHPND outbreaks reached USD 1.8 million for the monodon shrimp assuming 38% of production was from semi intensive and 63% extensive culture. In the case of the vannamei shrimp, it was USD 8 million as more ponds (52%) are semi-intensive. In the case of WSSV, we used 2% loss per day over a 110-day post stocking period. The disease caused losses of USD 2.3 million/year for the monodon shrimp and USD 5.6 million/year for the vannamei shrimp.”

Andy added that calculating losses from EHP outbreaks is more complicated as a high spore infection in the hepatopancreas affects the digestive capacity of the shrimp. “So we looked at a pond with a typical production of 110 days cycle harvesting 12 tonnes/ha of 18 g shrimp. With EHP, the growth ended at 12 g and production was reduced to around 9 tonnes/ha. The value was only USD 3.5/kg but at that point the production costs were around USD 4/kg. The loss was calculated at USD 32,000/ha. When we apply these figures in Thailand where a recent survey suggested that 49% of ponds are infected, the economic loss is USD 76.4 million/year.”

The message was: “Collectively, all these diseases are having an impact on shaping aquaculture growth and production in Asia. There are two different investment models, the first being low intensity production with low biosecurity meaning there are high probabilities of disease outbreaks but losses are generally low when they occur. In contrast, high intensity production with high biosecurity means lower probability of outbreaks, but when outbreaks do occur, the losses are very high.”

Living with Current Shrimp Disease Threats in Asia

In her presentation, **Dr Kallaya Sritunyalucksana**, Principal Researcher, National Center for Genetic Engineering & Biotechnology (BIOTEC), Thailand focused on the latest findings on the relationship between EMS and AHPND and what is known on EHP.

AHPND

Work at BIOTEC showed that the virulence mechanism of AHPND-inducing *Vibrio parahaemolyticus* (VP_{AHPND}) and that AHPND pathology in shrimp is caused by binary toxins called AHPND-ToxA (12.7 kDa) and -ToxB (50.1kDa) released from the bacteria colonising the shrimp stomach. The AP4 PCR detection method designed based on toxin information with highest sensitivity and specificity is now recommended to be used to ensure that post larvae and live feed are free of VP_{AHPND} and to monitor shrimp during the culture period.



Kallaya Sritunyalucksana

“Next is the cohort epidemiological study of 200 pre-selected ponds from EMS/AHPND areas conducted with Dr Varin Tanasomwang, Senior Expert at the Department of Fisheries. Ponds with early mortality (<35 days) were sampled immediately and sent for multiple analyses. All other ponds that showed no mortality before 35 days were sampled during 35-40 days and sent for the same multiple analyses. Only 30 ponds had EMS and in these only 40% was confirmed by PCR and 25% by histology

that the cause of mortality was AHPND. These results suggest that AHPND is only part of EMS, i.e. AHPND does not equal to EMS. In addition, we also found that a low level of toxin or low concentration of VP_{AHPND} did not cause cell sloughing, but collapsed epithelial of hepatopancreatic cells. The results from this study lead us to consider in redefining the case definition of AHPND.

“Recently, we found one VP isolate harbouring plasmid from Vietnam and one from Thailand that can kill shrimp, but not with AHPND. The take-home message is that there is a different mechanism killing shrimp and is not through PirA/B-like toxin.”

Kallaya also described the synergistic effects of the bacteria *Shewanella*, which has been co-isolated with VP_{AHPND} from the AHPND shrimp. Immersion with single bacteria of *Shewanella* at 10⁴ CFU/mL or 10³ CFU/mL VP_{AHPND} can cause mortality at 45% and 20%, respectively. Combination of both bacteria with the dose indicated can cause 100% mortality and suggested the synergistic effect of both bacteria.

“We conclude that there is a range of bacterial VP isolates of variable virulence. Some produce PirA/B-like toxins but vary in virulence that needs to be explained. Some produce no PirA/B-like toxins nor cause AHPND pathology but still cause significant mortality. We have more questions. Is virulence based on plasmid genes only? What about the prevalence of AHPND partners.”

EHP

The transmission of EHP directly from shrimp to shrimp by cannibalism and cohabitation, makes its control difficult. In the Thai cohort study, there is a high incidence of EHP in shrimp ponds and higher prevalence in the pond that reported no shrimp mortality before 35 days.

Kallaya shared results from a CP study which showed infections of EHP were not correlated with survival, but with weight, average daily weight gain (ADG) and size variation (% CV).

“On the link between EHP and white faeces syndrome (WFS), our laboratory trial showed that shrimp fed with EHP infected hepatopancreatic tissue and became EHP-infected did not exhibit symptoms of WFS even after 30 days. We conclude that EHP is not the direct cause of WFS. Our cohabitation studies confirmed horizontal transmission of EHP.

“On detection of EHP, the recommendation is to use the SPW-PCR method rather than a 16s rRNA-PCR method. This is because we have found the possibility of cross-reactions of 16s rRNA primers to other microsporidian in other aquatic species, not in shrimp. We have also developed the simple spore staining method with 2% aqueous Phloxine B to be used in the field. Lately, we have developed the method to activate the polar tube extrusion from an EHP spore, which is a way to infect the host cell. An on-going study is to try inhibit this process.”

Going forward, Kallaya said, “With the genome sequence of EHP in cooperation with Exeter University and Cefas, UK, we can understand better the virulence mechanism of EHP. We are now

working on the control of EHP based on the virulence genes we found from the EHP genome information.”

EMS/AHPND: The Complete Story? From Mechanism of Action to Prevention



Ung Eng Huan

Ung Eng Huan, Chief Technology Officer, Biovalence Sdn Bhd discussed on three aspects of AHPND. The first is the cause of blebbing cell sloughing and collapsed tubules and the second, the differences in virulence of the AHPND-causing bacteria, pir A/B PCR+ strains of *Vibrio parahaemolyticus* (Vp) and their association while the third is about how easy other *Vibrio* species could acquire the toxin-carrying genes.

“In the early days of AHPND, we found that with more severity of the disease, there would be more sloughing of the hepatopancreatic cells. We decided to look at the molecular mechanism of blebs. Blebs can only be seen through transmission electron microscopy (TEM) and during a small window of 45 minutes. The presence of zonal occludens toxin (ZOT) explains why cell sloughing of intact hepatopancreatic tubular cells occurs into the lumen during the early onset of the disease. ZOT acts by attacking the tight intercellular junctions.

“We also know that Vp bacterial strains are different from each other and that virulence may vary. We wanted to understand the level of virulence. Prophage elements linked toxic genes in strains of Vp are capable of producing histologically confirmed signs of the disease linked to toxic genes. These virulence/toxin genes were conferred by a lysogenic phage. ZOT on prophage 3 showed higher virulence than ZOT on prophage 2 but together, the virulence multiplies,” added Ung.

Bioinformatic evidence first presented during DAA9 in Ho Chi Minh City also showed that ZOT may have originally come from a species of *Shewanella* and that the presence of prophage elements is evidence of how bacteriophages were historically involved in the transduction.

“We have also carried out co-incubation experiments to show that a strain of pir A/B+ *V. parahaemolyticus* was able to quickly and easily transmit pirA/B to a strain of non-pirA/B *V. alginolyticus*, which explains recent reports of other *Vibrio* found with these genes. The danger is that sooner or later, other compatible *Vibrio* species may also become weaponized by simple conjugation via Type 3 secretion systems (T3SS). This may offer the potential for other compatible species that have other toxins to act in synergy with pirA/B or ZOT and give rise to an even more virulent pathogen. There are now also reports of pirA/B-containing strains that do not show cell sloughing despite mortalities being observed. We suggest investigating if ZOT is also present in these strains because these might be caused by just pirA/B toxicosis acting independently.

Health Management for Profitable and Safe Aquaculture Production

Many of the emerging diseases in shrimp are not fully understood. Often there is little that can be done at the therapeutic level when there is an outbreak. Industry should consider preventive or prophylactic measures to secure their business. With an extensive background in running a health management program at a Mediterranean fish farm, **Dr Maria Mercè Isern i Subich**,

Business Development Manager, Aquaculture Health, Nutriad International NV, Belgium gave her take on similar approaches in Asia's shrimp farming business.

Health management, aimed at disease prevention and avoiding the costs of disease and corrective measures is becoming a must for the shrimp industry to maintain its sustainable and profitable growth," said Maria Mercè. "The set-up of proper strategies needs to involve administrations, governments, academics and researchers, suppliers and then industry and farmers."

There is a need to establish the status of the epidemiological unit for each specific disease of interest comprising historical data and surveillance. "We never speak that 100% an outbreak will not happen. When we are not ready, this will happen. We are balancing risks of doing or not doing, having or not having a program. This is the hidden part of biosecurity."



Maria Mercè Isem i Subich

In aquaculture, few are able to work in closed environments. "We may think that we are isolated but are actually not. The farmer should ask how will production affect the environment and how will the environment impact the farm. A good example is control of sea lice in Norway. This shows organization at the farm, zone, area, region and country level. Here the farms in 550 sites with 8 cages/site submit data on prevalence of sea lice in salmon and 10 fish from four cages at each site are counted each week for sea lice. With the years, strategies were developed into biological, mechanical plans to remove the sea lice menace from farms favouring biological treatments and working to reduce optimized medical treatments as the latest solution."

How does this apply to shrimp farming? Maria Mercè said that this starts with the control and optimization of conditions for the shrimp (SPF/SPR, age and husbandry, feeding and optimal nutrition) and control of environmental conditions. "Something to learn is how the fish industry is very concerned with fallowing, not because they want to do this but because there is a need. Disease surveillances and monitoring include all mechanisms to know the health situation of the farm. For example, disease monitoring and checking for lipid droplets in the hepatopancreas as signs of a healthy animal and early detection of pathological changes.

"We cannot manage what we do not know we have and we will not find it if we do not look for it. Surveillances and monitoring will allow to implement tailor made procedures and strategies for the farm to avoid the consequences of disease," concluded Maria Mercè.

Sanitary Status and Genetics as Part of a Biosecurity Strategy'

Dr Victoria Alday-Sanz, Director of Biosecurity and Genetics, National Aquaculture Group (NAQUA), Saudi Arabia, started her presentation by dispersing some confusion in the industry on the concept of biosecurity. Her message is that biosecurity is a tool for sustainability.

"The objective of biosecurity is to reduce the economic impact of diseases. Biosecurity is more than activities in the farm to prevent, control and/or manage disease risks such as PCR, disinfections etc. Biosecurity includes the selection of animals depending on the culture system. We can categorise these for health status or genetic characteristics.



Victoria Alday-Sanz

specific pathogen resistance (SPR) and specific pathogen tolerant (SPT)."

SPR means that the animal cannot be infected with a particular pathogen but not with a range of pathogens. SPT means that the animal can get infected but will not develop the disease or will develop a disease to a certain extent depending on environment and culture conditions.

"The message is that when the farm is making a decision on its biosecurity level, it needs to look at health status and genetic characteristics of the animal and to consider history and culture conditions."

SPF is a health status

Victoria noted that "SPF refers to the sanitary status of a stock. It is not necessarily free from all pathogens, and not simply PCR negative. SPF stocks are free of certain pathogens regardless of its tolerance, resistance and susceptibility to any pathogen. SPF animals come from a population which has been tested negative for pathogens for at least 2 years with a surveillance program in place, raised in highly biosecure facilities, including enclosed water treated environments, following biosecure management measures, and fed biosecure feeds.

"It is not necessary that SPF animals are pathogen free, more sensitive to pathogens, more resistant or tolerant to pathogens, inbred or have lower genetic diversity and better growth. SPF is not heritable and the offspring loses the SPF status when exposed to lower biosecurity level."

Victoria added, "We work with SPF because infections are a physiological cost affecting productivity and lower performance of shrimp. We need SPF to be used for genetic improvement programs, research and is fundamental for international trade. Imagine the scenario if we had been moving around non-SPF shrimp?"

Pathogens and SPF

The introduction of SPF stocks has completely changed the industry in Asia. However, there is no consensus on what pathogens define SPF and it is flexible. It is clear that SPF animals are not free from all pathogens, and not simply PCR negative. Although SPF programs should target all known pathogens, i.e. OIE listed as well as other known pathogens.

"The specific pathogens are defined by the supplier and the client. Some use OIE listed pathogens but OIE is not dynamic enough. It only listed AHPND in 2016 and has yet to look at EHP. DOF Thailand listed only OIE systemic viruses, which include WSSV, IHNV, YHV, IMNV and TSV. India requires SPF for all OIE listed pathogens and Egypt, all *Vibrio* bacteria and fungi but no virus."

She gave details on the surveillance program such as techniques (PCR and histology) and frequency, internal and external certification, participation in international PCR lab ring test and latest tests performed. The findings of an FAO

Expert Workshop on SPF, SPR and SPT: a Need for International Technical Guidelines will be released in 2017. These will provide a better understanding for the industry.

Genetic characteristics

“The concepts represented by the acronyms SPF, SPR and SPT have been used in a rather confusing manner over the last few years. SPR and SPT refer to their genetic characteristics that allow them to be resistant to infection to a particular pathogen or tolerant to the development of the disease caused by a particular pathogen.

These are genetic characteristics regardless of their sanitary status, whether the stocks are infected or not. In other words, stocks can be both SPF and SPR/SPT. SPR/SPT may, in some cases, have the drawback of lower performance when compared to other stocks. While resistance, tolerance, susceptibility or higher productivity is a choice based on the biosecurity risk of the farm that will use them, the sanitary status should not be a choice. Why would anyone want to stock (invest) in infected animals?

Developing WSSV SPT+SPF

During WSSV outbreaks, some shrimp producers, such as those in Thailand, chose the SPF route and adapted culture conditions with high biosecurity. However, Ecuador and Latin America simply coped with WSSV in low biosecurity ponds and took a longer time to recover.

“In a natural way, WSSV was present in ponds in Latin America for almost 15 years and shrimp developed tolerance against WSSV. But for the future, we need to be ready as the impact will be high if there is an epidemic such as EMS entering vertically into the system. In Latin America, SPF was not adopted because industry associated it to disease susceptibility.”

Victoria detailed efforts in Ecuador and Nicaragua in 2010 to clean up WSSV SPT shrimp to be SPF as well. “We started by monitoring pathogens in 23-30 g shrimp from ponds with no biosecurity, over a broad geographical spread for maximum genetic diversity and from ponds with low performance. We then proceeded with nation-wide surveillance using the OIE listed pathogens in 2010 and added in EHP, PvNv, and Streptococcus. Results showed that infections present included WSSV, IHNV and NHP, but TSV, IMNV, YHV/GAV, BP, PvNv and EHP were not detected. The goal was to exclude these three pathogens in the SPF development.

“Seventy-five shrimp were selected from each pond. These were from ponds with good productivity, low disease prevalence and good appearance. This was repeated for 3 generations. Individual testing was conducted on shrimp after spawning and stressed at 22-24 °C for 48 hours to replicate WSSV. NHP was eradicated with antibiotic therapy of broodstock and antibiotic egg washing. SPF was confirmed by the Department of Infectious Diseases, University of Zaragoza in Spain, an EU-recognised pathology laboratory.

Does SPF bring advantages to production? “A new term for this process is ‘Reverse SPF’. These WSSV SPT/SPR SPF shrimp combine sanitary and genetic characteristics. Assessments on mortality post ablation and during production showed lower mortality rates. In an intensive system, survival was better than SPF shrimp imported from the US. These SPF+WSSV SPT vannamei shrimp were used to recover production in NAQUA in Saudi Arabia which was devastated in 2013 due to WSSV. Production picked up to 22,000 tonnes in 2015, higher than any prior production.”

The message was that SPF animals are fundamental to any production unit and genetic characteristics in terms of response to pathogens need to be suitable to culture conditions.

Adapting to a New Reality: Shrimp Nurseries



Jesper H. Clausen

According to **Dr Jesper H. Clausen**, Senior Assistant Product Manager, Farm and Feedmill, INVE Aquaculture, Thailand, there is a move towards nurseries due to the increased occurrence of diseases in shrimp farming today. “Diseases are here to stay, and we, the industry, have to manage production to minimize the impact from these diseases. This is the new reality and the industry does not have a choice except to adapt. Shrimp nurseries are not new and used to be common in the industry in Latin America and Asia before Asia’s shift to white leg shrimp. However, farms then went on to direct stocking from hatchery to grow-out in ponds. Today, the nursery phase is reappearing in Asia.

“Nurseries were used to speed up growth before the grow-out cycle and this remains a valid reason for nurseries in Latin America. In South Korea, nurseries are used to give a head start in shrimp farming with the nursery cycle over the winter months. This emerging trend in Asia is more so because of diseases in the early stages of shrimp growth. Today, we also talk about the need for robustness rather than just fast growth as the latter will not work if shrimp mortality is high during the early stages.”

Among the diseases faced by the industry today are *Enterocytozoon hepatopenaei* (EHP), acute hepatopancreatic necrosis disease (EMS/AHPND) and white faeces syndrome. In the case of *Vibrio* causing AHPND, the use of antimicrobials is now a major concern. According to Jesper, a few years ago, the industry had eliminated the use of antimicrobials from many Southeast Asian countries; but in recent years, there has been a sharp rise in EU rejections and detentions with Japanese and USDA data also showing similar trends. These were often due to antimicrobial residues in shrimp products from countries with EMS/AHPND outbreaks.

Changing protocols

“There are no silver bullets against diseases and farms will need to work together to adopt management practices towards an economically sustainable direction,” said Jesper. The presentation included descriptions of some nursery systems which have been developed recently as well as some commercial trials. More details are available in his article in issue July/August Aqua Culture Asia Pacific 2016, p. 15-18.

“A benefit which is sometimes forgotten is that nurseries are an extra checkpoint where shrimp can be checked for diseases before stocking into ponds. I advocate that the new normal should be well managed nursery systems using high quality diets, feed additives and probiotics for a healthy environment. These include elevated doses of nutraceuticals and specific immunostimulants, vitamins, lipids, pigments and nucleotides to support osmoregulation, growth and other vital processes during normal and stressful conditions,” added Jesper.

With a 15% replacement of quality feed during the PL15-PL27 period, Jesper showed that post larvae growth was faster with higher biomass. Nurseries with low water exchange will require probiotics and the tip given was that farmers should calculate cost based on CFU count in the product and not based on the actual volume of the product.

“With regard to stocking density, traditionally it is 2-5 PL/L and feeding using traditional starter diets similar to during the early days in the ponds. Moving forward, using a high quality product nursery protocol, it is possible to increase stocking density to 30-35 PL/L. In Vietnam, INVE has shown that this can save the farmer 14% in overall cost at the end of the nursery cycle.”

“It is not one size fits all for the nursery protocols and for the investment of the farmer. A tailor-made nursery protocol should be used for each situation. Farmers in Thailand’s southern provinces are using the shrimp nursery with success and are shortening the cycle for the nursery phase.”

Supply Chain and Ingredient Excellence: Foundations of a Stable Shrimp Industry

“Along the supply chain, there are several expectations. Farmers want healthy shrimp, fast growth and excellent growth performance through standardized feed and stable feed prices. Because marine ingredients are high, upstream in the supply chain, or any weakness will affect production. The inclusion rate of fish meal is now lower, but as shrimp farmers regard fish meal as the gold standard in feeds, it is paramount to deliver their expectations with shrimp diets,” said the SPF-Diana Aquativ team of **Dr Philippe Sourd**, Technical and Sales Director and **Vincent Percier**, General Manager in their joint presentation.

“As shrimp aquaculture expands, we see that the weak links are in ingredient quality, standardization and consistency. Beyond the nutritional profiles of the ingredients, there are other issues such as supply, sourcing, logistics, price, quality, specifications, certifications, packaging, physical aspect and seasonality. We need to address all these if we want to build a stable industry. This consistency only can allow predictability in farming performances,” said Philippe.

Variability with fish meals

Fish meal has played a role in shaping the industry, even though supply is unstable and prices are constantly changing; from USD 1.3/kg to USD 2.3/kg over a 4-year period.

The team discussed findings from a 2015-2016 market survey on fish meals sampled worldwide. The analytical profiles of these ingredients were followed by feed trials. Some 37 fish meals comprising three types of fish meals: super prime, standard grade and co-products from global sources were analysed.

“Are product specifications enough to select an ingredient? As expected the protein and ash content are statistically different among groups of fish meals. The surprise is with soluble proteins



Philippe Sourd

Vincent Percier

and small peptides which prove a lot more variable within and between fish meal grades. Processes drive product specifications. In the case of biogenic amines, which are sometimes used to grade fish meals, we saw that amounts did not differ statistically.”

Philippe also reported on the variations in protein (69-75%), soluble protein (3-14%) and peptide content between batches of super prime anchovy fish meal from the same origin. He said that the variation is a challenge for feed formulators tasked with delivering the same diet with the same growth performance from this high grade fish meal. More details on this are available in an article published in the July/August Aqua Culture Asia Pacific 2016 issue, p44-47.

Marine meals in Southeast Asia

“Results on analyses of shrimp and squid meals indicated that shrimp meal appears to be well standardized but with low digestibility. However, squid meal which shrimp farmers consider as a desirable ingredient in shrimp feeds, vary with different protein levels as well as digestibility. The product with the highest protein did not show the highest digestibility,” said Philippe. “In the case of local fish meals, most have 60% crude protein. Co-product fish meal is frequently used in SEA. Some contain high levels of soluble protein and peptides. Freshness is sometimes an issue with high putrescine levels. Deviations in digestibility are high; hence again the importance of a standardized process.”

Drivers of standardization

“The salmon silages from Alaska, Australia, Scotland, Denmark and Norway showed different levels of protein and soluble protein contents. All these clearly indicate that product specifications alone do not define a product value. The raw material selection and the process itself are crucial drivers of a product’s profile.

“For each marine ingredient, processing can influence product standardization. Factors of influence range from species selection (mono or mixed species), tissues (whole fish or viscera or both) and in fish meal, drying temperatures or whether solubles are added into the meal.

“Achieving both standardization and consistency are not impossible. Among 107 batches of a tuna fish hydrolysate, the batch variation was 22.08 ± 0.43 for crude protein, but most importantly, soluble protein and peptide profiles remained exactly the same. Our belief is that by adding such highly standardized raw material into the feed recipe, we will be able to offset any variability found in marine or plant ingredients.”

Feed trials

A series of 8-week feed trials with sea bass allowed the ranking and benchmarking of 18 fish meals (prime, standard and co-product) from Mexico, Denmark, Turkey, Peru, Norway, Mauritius, India, Argentina and Ecuador. These were added into plant meal diets and scored versus a positive control (100% prime fish meal diet) and a negative control (100% plant meal diet).

In another group of 1.5-month feed trials, two fish meals (one super prime and one co-product) were blended and used in several recipes at inclusions of 25, 20, 15 and 12.5% while a hydrolysate was added at a 1:2.5 proportion.

“From the first series of trials, we concluded that some fish meals can actually bring down growth performance, and that some standard fish meals could perform better than prime grades.

“In the second series of tests we verified that using a low performing co-product fish meal in diets meant that growth performance went down; and the more co-product fish meal in the diets, the more growth performance deteriorated. However,

the addition of hydrolysates could restore growth up to the level of growth obtained in the super prime fish meal fed group. This showed that hydrolysate can truly be a tool to offset performance variations driven by fish meal grades, and provide an efficient solution for formulators who have to compose with their raw material basket,” concluded Philippe.

Consistent and stable supply

On the challenges for an ingredient producer, Vincent noted, “Working on hydrolysates require probably the highest level of product standards. Our contribution to the supply chain is a guarantee on exactly the same peptide profile for each batch. To deliver a constant supply of a standard product, we need to have the same species and same tissues at a level of unquestionable freshness. Of course our price has to be stable too.”

The next step is working alongside suppliers of raw materials and to maintain stable pricing. “For this, it is essential to have them on board,” said Vincent. “We have learnt that sustaining consistency and stability is a business. Customers working with us realise that we can supply consistent ingredients and have shared values for the ingredient industry. The source owner and ingredient manufacturers are the beginning of the stability chain and we cannot afford to have gaps. A stable chain allows the industry to have a stronger control of the value chain.

“Also, the shared objective is not enough. We need to communicate on what the industry needs. Then the next step, which may take two or more years, is certification. This is the road map for this industry.”

Dealing with Deadly Diseases: Reviewing Nutritional Solutions Available to the Shrimp Industry

There is a need to change the perception that shrimp farming is a disease-prone industry. Nutritional interventions could possibly help but prior to this, it is important to understand crustacean immunity. In his review on what is available in the scientific domain, **Dr Kabir Chowdhury**, Global Product Manager, Jefe, Canada discussed the unknowns in shrimp immunity followed by some action mechanisms comprising solutions and additives, how and when to use these products as well as the environmental and economical consequences of their use.

“As most pathogens are already prevalent in apparently healthy populations, disease outbreaks are usually a result of massive amplifications following exposure to various forms of environmental and physiological stress. Stressors can include handling, spawning, poor water quality or abrupt changes in temperature and salinity,” explained Kabir.

Knowing the unknowns

“Crustaceans rely on a nonspecific immune mechanism to prevent entry and spread of pathogens. When the mechanism fails, crustaceans immediately initiate multiple innate immune responses to defend against the pathogens. Both humoral (involving cell free components of the haemolymph) and cellular (components by haemocytes) components work together in case of a pathogenic outbreak.

“There is a diverse array of humoral immune responses include clotting cascades, antioxidant defence enzymes such as superoxide dismutase, peroxidase, catalase and nitric oxide synthase, defensive



Kabir Chowdhury

enzymes such as lysozyme, acid and alkaline phosphatases, and antimicrobial peptides. The cellular component involves phagocytosis, apoptosis and RNA interference. One of the most effective immune mechanisms in invertebrates is the cellular melanotic encapsulation. A lot of excellent papers are describing this prophenoloxidase (proPo) system,” said Kabir.

“Recently, a report on the trained immunity in invertebrates during a transgenerational study using *Artemia* as a model was published by Norouzitallab et al. (2016). In this study, the authors exposed successive generations to *Vibrio campbelli* and then exposed all three generations simultaneously to the *Vibrio*. This work suggested that innate immune responses in invertebrates have the capacity to be trained, and epigenetic reprogramming of (selected) innate immune effectors is likely to have central place in the mechanisms leading to trained immunity.”

Feed based solutions

Several feed additive based solutions to prevent disease occurrence or to improve non-specific immune responses are available worldwide. Kabir divided the solutions into inorganics and organics. Organic solutions include but are not limited to pro- and pre-biotics, essential oils, components from macro- and micro-algae or single cell proteins, carotenoids, organic acids and their salts, and enzymes. He explained how metallo-protease work in the proPo system.

“Inorganic solutions include several divalent metallic compounds and their chelated products with amino acids such as lysine and methionine as well as carotenoids. Lin et al. (2013) looked at how dietary sources such as zinc methionine (ZnMet), zinc lysine (ZnLys), zinc glycine (ZnGly) and zinc sulphate ($ZnSO_4 \cdot H_2O$) affect growth and immune parameters for *Litopenaeus vannamei*. Shrimp fed diets with organic zinc supplementation produced significantly higher growth, survival and immune parameters than $ZnSO_4$ treatment.”

In 2013, Zhang et al. showed that shrimp fed six dietary astaxanthin levels gave higher survival than the control after a low dissolved oxygen stressor. According to Xie et al. (2015), 2.29–2.34% of proline in the low fish meal diet could improve anti-oxidative capacity, immune response, NH_3 stress tolerance of *L. vannamei*, and proline may be a conditionally essential amino acid for the vannamei shrimp.

The potential of membrane active peptides (MAP) was also described. Koh et al. reported a long list of plant extracts, essential oils, bioactive metabolites, plant exudates and fungal extracts, inhibiting quorum sensing of various bacterial species.

Interest in enzymatic function for protecting against microbial infection has intensified in recent years. Some of these include halogenated furanones (from seaweeds), synthetic auto-inducing peptides and paraoxonase enzymes (PONs). Kabir discussed the research by Song et al. (2016) on the effect of a dietary protease-complex on growth performance, body composition, digestive and immune enzyme activity of *L. vannamei* and its resistance to a pathogenic *Vibrio parahaemolyticus*. A high fish meal diet containing 200 g/kg fish meal was the positive control and a low fish meal diet containing 100 g/kg fish meal, the negative control. For the other treatments, the low fish meal diets were supplemented with graded level of a protease-complex (125, 150 and 175 mg/kg, respectively). Total superoxide dismutase and polyphenol oxidase contents in both serum and hepatopancreas were higher and serum malondialdehyde content and the cumulative mortality during disease challenge tests were lower for the diets containing the protease-complex than those fed the low fish meal diets ($P < 0.05$), with no difference with those fed the high fish meal diets.

“The new paradigm will be bacteriophages and in the future, we will hear more on the use of bacteriophages against diseases. However, an issue with bacteriophages is that they need to be changed every 2-3 years as they lose efficacy.”

Disease Mitigation via Feeds and Feeding: Feed Industry Perspectives



Daranee Sookying

How can we use our nutritional knowledge and feed management practices to mitigate diseases in shrimp? In her presentation, **Dr Daranee Sookying**, Regional Nutritionist, Aquaculture, DSM Nutritional Products Ltd, Thailand discussed options through the eyes of the feed industry and shrimp farmer. She said that farmers have accepted the fact that poor nutrition and poor feeding practices may lead to a reduced immune system response and lower the ability of shrimp to resist disease. However, at the same time, the demand is for reasonable and consistent feed performance in terms of ADG and FCR and good quality harvest at reasonable feed costs.

“We need to optimize feed performance away from just formulation. We need to look at impact of feeds which we can control. By reducing protein and excess phosphorus, we can reduce the organic load in pond water. This then leads to less water quality problems, less stress, lower risk of disease outbreak and better performance.”

“When we replace fish meal with plant meals, we then need to improve feed digestibility, attractability and water stability. Use of enzymes, although more common in fish feeds than in shrimp feeds are options. Phytase increases available phosphorus and reduces environmental impact. Xylanase breaks down cell walls and protease can increase nutrient availability and minimize organic wastes. Sustainable marine proteins serve as feed stimulants and attractants. The soluble protein requirement is 8% for vannamei shrimp feeds and 10-12% in feeds for the monodon shrimp. Fast growing shrimp requires highly available nutrients.”

According to Daranee, black spot or black gill disease, cramped muscle syndrome, soft-shell syndrome, blue shrimp and red diseases are nutritionally linked. Specific nutrients such as Vitamin C supplementation help with black gill disease. Calcium and phosphorus supplementation help with cramped muscles. “It may be possible that soft-shell syndrome could be due to rancid/low quality feed and high energy feeds and mineral supplementation can help. Blue shrimp disease or blue shell syndrome is caused by low oxygen in ponds or low level of astaxanthin in feed. Astaxanthin improves biological functions and improves survival, growth and stress resistance in shrimp.”

“Vitamins are known to enhance disease resistance. Today, the vitamin levels in feeds are at less than optimal requirement levels. Superior dietary supplementation levels of certain vitamins do provide additional value. Dietary inclusion of vitamin C is required for growth and development, reproduction, resistance to disease, enhanced immune system response, stress reduction, wound healing and enhancement of antioxidant properties. Elevated dietary levels of vitamin C have been shown to improve resistance to diseases, salinity shock and other stresses. In the case of vitamin E, the recommended level is 100 ppm. Higher supplementation levels are related to dietary lipid level or stress.”



Managing EHP and Production Planning to Maximize Output

Dr Celia R. Lavilla-Pitogo, iAqua Malaysia, is a scientist with extensive experience on diseases affecting farmed shrimp and fish. Her most recent work focusses on the management of the microsporidia *Enterocytozoon hepatopenaei* (EHP) at a farm in Malaysia. Celia started with an explanation on how the microsporidian infects shrimp.



Celia R. Lavilla-Pitogo

“The less than 1 μm spore infects the shrimp when the missile-like organelle releases the polar tubule. Once in the host, it replicates. Each infected shrimp in the farm contributes infectious spores into the environment through its faeces. Microsporidian spores are resistant to drying and routine chlorination levels. Disinfection to deactivate spores requires out-of-the-normal biosecurity protocols like exposure to high or low pH.”

“The missile-like polar tubule has a single-release mechanism and failure to inject itself into a host subsequently deactivates the spore. We can prevent infections if we can find means to deactivate the missile organelle. Until then, we have to focus on diagnosing the severity of infections and how to manage production around EHP infections,” explained Celia.

Wet mount microscopy

EHP can be detected by polymerase chain reaction (PCR), the kits of which are now commercially available. However, detection of EHP by PCR provides limited information on how best to manage the stocks after detection. “We need to know the severity of infection. I recommend wet mount microscopy of fresh hepatopancreas samples which allow grading of severity of infection. Histology is also a good tool to confirm severity grading and location of affected cells of the hepatopancreas,” said Celia.

In describing the steps for wet mount microscopy, Celia emphasised on the need to include the central part of the hepatopancreas to see spores inside infected cells. Once infected cells are located, the severity of infection can be determined. This diagnosis can be partnered with a real time PCR to validate microscopic grading of severity of infection.

“Under farm conditions the number of infectious spores can be minimised by thorough disinfection of farming premises with the application of high pH 12 using burnt lime or low pH 4 using HCl. The focal points for disinfection can be the seams of pond liners. In hatcheries, prevention of direct contact between eggs and broodstock faeces is a key strategy but can be difficult and laborious, such as using condoms on female broodstock. This was done to prevent transfer of monodon baculovirus (MBV) several years ago.”

Production planning

“At the farm, infections with EHP cause a wide variation in sizes despite shrimp eating well. We graded the severity of infections; and linked these to critical factors such as the average body weight (ABW), average daily growth (ADG) and feed conversion ratio (FCR). We then made decisions on how to manage stocks in affected ponds: whether to continue, harvest, or abort the crop,” Celia continued. “Usually, when grade 3 severity and above is observed and growth has plateaued, the farm prepares to abort or harvest to hasten pond turnaround instead of waiting for larger shrimp sizes. However, decisions will need to depend on markets too, based on the minimum ABW that has a value. In Malaysia, for example, shrimp below 7 g has no value. Production planning based on the EHP severity and observed abnormalities will be part of the new normal in shrimp farming.”

The take-home message was, “There should be timely communication between production staff and the laboratory technicians. This is currently a weak link in our shrimp industry.”

Penaeus monodon in MRAS: A Cost-Effective Model For Controlled Production in China

In this presentation, **Robby Mort**, Aquaculture Manager, RADAQUA, Australia shared his experiences in running a cost effective managed recirculating aquaculture system (MRAS), which the company designed and constructed in Fujian Province, China. The commercial trial for the production of *Penaeus monodon* ran from December 2015 to harvesting in February 2016.

In his introduction to RAS, Robby listed reasons for such controlled production models. RAS can be located anywhere, close to markets or for food security reasons. Legislation is also a reason for RAS such as in Australia with environmental compliance regulations limiting open culture systems.

“These are engineered systems where water coming in is filtered and there is continuous water treatment. We can control parameters such as pH and temperature, even to the point of adjusting photoperiod. RAS gives us high unit output per unit of land, and minimises business risk without any production interruption. Biosecurity allows a high degree of control over external risks,” explained Robby.



Robby Mort

The system in China uses standard round HDPE 65 m³ fish culture tanks. The culture period was divided into specific phases. The nursery phase uses post larvae PL25 in 4 m diameter tanks for a duration of 5 weeks. Aeration was heavy and probiotics enhanced algae and bacteria blooms. The grow-out phase of 6 months was spread over the winter season. Robby said that when they were

producing without interruption, shrimp farms in the surrounding areas remained fallow over the winter months.

Uninterrupted production

“We graded shrimp regularly and adjusted biomass to manage stocking densities and aggression. Shrimp were fed 100% commercial diets. As shrimp reached 30 g, we carried out partial harvests. The harvest was planned in February 2016 to get the best prices and shrimp were sold at average prices of 200 RMB/kg (USD 29.5/kg) for live product through niche buyers. The total production was 2 kg/m² which was equivalent to 20 tonnes/ha. It helped that the location of this facility was between two tier 1 cities. The choice on location was a business decision of the client. Our advantage was that we could produce large monodon shrimp of size U10 at 10-15/kg. We had little competition as other monodon shrimp producers had disease issues during the winter months.

In terms of productivity, culturing shrimp in MRAS has an advantage over traditional farming in open pond systems where productivity is 10 tonnes/ha in Australian shrimp ponds and 5 tonnes/ha in Vietnam.

The limitations are the high capital outlay. The staff required should have more than the average technical background. Despite relatively low yields, the commercial trial demonstrated that there is potential to use MRAS for the commercial culture of *P. monodon* in China. In a normal scenario for RAS, where the ratio of water: fish is 1:10 the profits should be better.

“There is a strong demand for biosecure shrimp production globally. RAS can be part of the new normal with the benefit of biosecurity for hatchery and advanced juvenile systems for the shrimp industry to get the existing pond culture off to a strong and healthy start.”

Proactive Management to Keep Disease Away 2.0: An Overview of Current Shrimp Farming Practices



Soraphat Panakorn

Soraphat Panakorn, Commercial Development Manager, Aquaculture Asia Pacific, Novozymes Biologicals, Thailand has visited shrimp farms in the region extensively and has seen how the industry, in particular, Thailand, is suffering with frequent outbreaks of early mortality syndrome and acute hepatopancreatic necrosis (EMS/AHPND) as well as white spot syndrome virus and white faeces disease. At TARS 2014, he presented on proactive management to keep disease

away. However, as diseases persist, Soraphat’s current focus is to get farmers to change mindset and farm management.

“Looking to the future, there is the possibility that shrimp farmers will continue to encounter new diseases while dealing with existing ones. Uncertainties and frequent crop losses from diseases are not sustainable for any industry. Following the phrase, ‘prevention is better than cure’, to survive in this industry, farmers will have to change their perceptions on the farming process.

“Today, the goal is to fight diseases. For this, sometimes the farmer needs to change the mindset. For each crop, he or she may need to think proactively. Farmers must know some basic

knowledge in shrimp farming. Being open, sharing knowledge among peers and accepting change are critical for success. But as each pond has its own features and conditions, the farmer should not copy outright practices but adapt to his own farm and pond conditions.”

Soraphat categorised target farmers as proactive management (PAM) farmers and listed the actions and practices they should follow. “The list is very long but important. It is only through proactive management that we can manage shrimp and avoid problems. Farmers should work at fine-tuning processes along the supply chain and understand that behaviour and physiological needs of the living shrimp will change with environmental conditions. In turn, there is the dynamics of climate, season and other natural conditions, which will impact shrimp farming.

In farm management, farmers need to understand the science and learn to effectively use many of the aquaculture inputs such as disinfectants, lime, microorganisms and feed additives. In outdoor ponds, they need to prepare for climate and seasonal changes. Knowing local and global market situations is important for any investor as well as the farmer.”

“Efficiency and adapting SOPs are critical and this is influenced by the social well-being of the farm employees. We should not move away from basic practices but continue to innovate and select the most effective techniques as standard protocols to avoid some common problems. The change is moving shrimp farming from an art to science with the implementation of ideas and modern practices.”

Hard Talk with shrimp farmers on business models and disease mitigation

The three invited farm CEOs not only come from diverse backgrounds but they also manage farming enterprises and business models of diverse sizes.

Abu Bakar Ibrahim, CEO of Malaysia’s Blue Archipelago Berhad (BAB), is responsible for the integrated operations of two farms, a processing plant and hatcheries. **Dr Manoj M. Sharma**, Director of M/S Mayank Aquaculture Pvt Ltd in Surat, Gujarat, began farming large sized monodon shrimp in four ponds in 2009. **Dr Surapol Pratuangtum** is CEO of Bang Go Farm in Suratthani, Thailand. He has been in the industry for more than 30 years and is President of the Thai Marine Shrimp Farmers Association.

Business models and competitive edge

BAB is a subsidiary of Khazanah Nasional, the strategic investment arm of the Malaysian government. Abu Bakar’s task is not just confined to its commercial operations but also to spearhead shrimp farming in Malaysia to attract future investments into this industry.

“I have two hats, the commercial one to make sure that my entity is viable, and a development hat to create a business model which incorporates scale from the beginning. Thus, we did not have the luxury of an organic growth. We need to show a sustainable model, particularly to the investment community, before we can attract the right kind of investments into the country,” said Abu Bakar.

“This puts significant pressure on myself and my management team to deliver a set of results that can demonstrate that scale helps to develop the industry. Setting up a farm of this scale has its challenges. A small farmer will be nimble and proactive. But without scale they will not have the support services and structure of organisation. There are pro and cons. In building scale, we have to look at all the components, and we need breadth and depth of knowledge to be sustainable.”

Manoj said, “I decided to go with the natural carrying capacity to produce large size shrimp. I studied this model very carefully and now advocate and educate my fellow farmers to adopt this model. I stock 20-25 PL/m² to produce 10-20 count shrimp. I think I have a sustainable model that others can adopt. I may never have made a lot of money but I have never lost a single crop. I do not produce more than 5-6 tonnes/ha.

“Big size shrimp is more profitable for me. For the last 18 years, the harvest is booked 2 months before harvest. I think it is a good combination of art and science to grow to this size and requires a lot of discipline. I know aquaculture is a complex and difficult science but it is important that this is made simple especially when you are part of a cluster,” added Manoj.

Surapol said that his business model is typical in the Thai shrimp farming industry. “It has been unfortunate that shrimp farming in Thailand is still in ‘ICU’ in 2015. Although my farm is 35 years old, it is still functioning. We have our ups and downs. I stopped farming for 2 years. However, farmers meet twice a month to brainstorm on how to farm shrimp again. At the moment, we can see that things are improving as we now have good quality post larvae. The bad news is that pathogens are around still.”

Experiences with diseases

BAB’s west coast farm was hit badly by EMS/AHPND in 2010, together with other farms in the area. Mitigation steps included lining ponds and trying different solutions.

“We did not have the flexibility of closing temporarily as we needed to sustain jobs. Fortunately, production continued at the new farm which is in an isolated location and has good water quality. At the end of 2014, we sat down and analysed. We brought in specific pathogen resistant (SPR) shrimp from Latin America and stocked them in the west coast farm. The shrimp were found to be resistant to WSSV and EMS.

"But just at the time we were back to the normal 70-80% survival, shrimp stopped growing due to EHP. In our east coast farm, we are able to plan production around EHP," added Abu Bakar.

Manoj recognised that diseases are bound to come but by managing the stress factor, Mayank Farm manages to keep diseases away. "As a group in Surat, we signed a protocol, whereby once the monsoon starts, we stop farming from February to March. We harvest before June/July. In this way, we save our ponds and the entire area. This has worked and we are 90% successful. My own pond management style is keeping the ponds in a high state of health.

"In Suratthani farms, WSSV is more serious but it does not mean that we have eradicated EMS. We have learnt that we cannot allow EMS or any disease to infect ponds. How do we control diseases? We now have the 3Cs program - first C is clean post larvae. We now have clean post larvae free from EHP and EMS. The second and third Cs are clean ponds and clean and clear water. The first C is the responsibility of the post larvae supplier and the second and third are my responsibility. I strongly believe that the transfer of infection is through the post larvae," highlighted Surapol.

When costs are rising

Since 2012, survival rates have dropped tremendously but production costs have gone up. The panellists were then asked their strategies to maintain margins.

"While costs have gone up, thankfully prices have gone up and margins have been good. Moving forward, our concern is to bring down costs across four areas: post larvae, feed, energy and labour. We know that labour in the Malaysian context is not coming down. Productivity needs to be tackled. Our subsidy for electricity might disappear soon. We can automate some of our processes. Fundamentally, our need is to get the best value post larvae and tackle cost of feeding the shrimp. We need to bring feed conversion ratio (FCR) to 1.1 to 1.3, which is the top of any farmer's agenda. We need to work with the feed industry to keep costs down," said Abu Bakar.

"Some 50-60% of cost is feed which has increased to IND 87/kg and the calculated FCR is more than 2, based on feed sales. The biggest cost saving is to increase feeding efficiency. The farmer needs to devise his own science to reduce costs. Fortunately, post larvae prices have remained the same," said Manoj.

Surapol believes that in Thailand, shrimp farming can remain profitable provided farms can control diseases. He reiterated that the critical factor is good quality post larvae together with good management practices. In intensive systems, survivals have improved to 70-80%. Concurring with Abu Bakar, Surapol said that control should be with costs for energy and labour.

"Thailand can recover but not to the high annual production of 600,000 tonnes as before. In 2016, we better keep this at 400,000 to 450,000 tonnes, at most," said Surapol.



Abu Bakar Ibrahim, CEO, Blue Archipelago Berhad (BAB), Malaysia.

Dr Surapol Pratuangtum, CEO, Bang Go Farm, Suratthani, Thailand.

Dr Manoj M. Sharma, Director, M/S Mayank Aquaculture Pvt Ltd, Surat, Gujarat, India.

Tars 2016 : Report on Roundtable Breakout Group Discussions

Pathways to Disease Mitigation via Genetics, Functional Feeds and Production Technology

Genetics

The costs of running a breeding program and level of technology are high resulting in a limited number of companies developing genetic programs. Increasing the number of multiplication centres could improve industry access to better genetics and provide more opportunities for localised breeding goals. Confusion exists over running a breeding program and selling broodstock.

Many companies sell broodstock without any real breeding program behind them. Copycat hatcheries buy broodstock from breeding programs and then breed to supply to farmers. The IP protection strategy used by breeding programs and companies increases the risk of inbreeding in copycat programs. Some traits are highly affected by inbreeding, and inbreeding depression can manifest quickly. Without IP protection, companies would not want to invest. Farmers will lose if the animals are too inbred.

In terms of genetics, SPF/SPR/High Health gets mixed up in the breeding program, due to complexities with breeding for disease resistance in many animals. However, selective breeding for resistance to WSSV and AHPND seems to work to a degree it has been very successful for TSV. SPF is a health strategy for identifying and maintaining animals that are free of specific disease(s). SPR is a product developed from any number of genetic tools (e.g. family selection, mass selection, and marker-assisted selection). The development of an SPR line is not related to or is dependent on SPF stocks; however, SPR stocks can also be SPF. While SPR and SPF are not directly related, they are not mutually exclusive.

Shrimp from a genetic line cannot be expected to perform equally well under every environment and different levels of management and production technology. On developing the breeding index to cover key traits, it is important to incorporate the economic values for each trait in the selection index. Unfortunately, the economic value of traits varies from country to country and among farms within a country, and on market conditions and operation costs (energy and feed costs). This is where multiplication centres can help the industry.

On fraud and verification, governments can be involved. In Hawaii, state officials take samples and have a chain of custody (University of Arizona), and issue health certificates - an important element for traceability.

To reach the desired performance, there should be some level of environmental control to allow the animal to reach its genetic potential. Growth as a trait tends not to show a strong genotype x environment interaction. Selection for growth in one environment tends to have a positive impact on growth in all, if not most environments. The farmer's expectations from a single genetic line from a breeding program should be realistic. Site- and country-specific lines will be extremely expensive. It is more cost-effective to use management and production technology. The more traits you pile onto a breeding program, the more expensive it becomes.

Functional Feeds

In Asia, currently few functional feeds are being sold or labeled as functional feeds. This does not imply that in Asia, there are no functional feeds in use or there is no R&D on functional feeds. In contrast, for salmon production in Europe, functional feeds have become more abundant than normal feeds where 60% of the feed sold is considered functional. In the Mediterranean, feeds have evolved towards functional claims, health, environmental and seasonal needs.

Specific and broad-spectrum functional feeds can address many health issues, many different life stages or different environments. However, specific functional feeds can be a logistics nightmare for the feedmill's production. How can a feedmill produce without sufficient demand or produce quickly in response to an urgent request from a farm to address a problem? Another observation is whether so many functional feed types can be produced and marketed to answer all the diverse systems used in Southeast Asia.

Weaknesses identified were a combination of measurability, predictability on performance, liability and trust. Cost-conscious farmers perceive functional feeds as more expensive than standard feeds. Feedmillers find it difficult to define their feeds as functional feeds or they may not want to claim their feeds as such because consistent performance is rare with the changing culture situations. At the farm level, what are the cost implications and are these costs justifiable? The evaluation is on the ROI (for farms and feedmills) as such feeds involve biological measurements.



Suggested KPIs can be survival, in the case of nursery feeds, or a very good proof of concept. With disease threats, there are too many additives in the market tempting farmers to “top dress” rather than wait for the feedmiller to suggest accordingly. The Asian farmer needs to be convinced of the efficiency of the functional feed, but they are warming up to the concept of functionality to solve and prevent health issues.

Clearly, there is a need to work towards the 3Cs (communicate, collaborate, and commit) i.e. better collaboration between solution providers. Communication between solution providers, feed producers and farmers, coupled with strong technical and educational support is critical. There should not be ‘black boxes’ as the industry wants to understand what they are buying. A wish list is for opinion leaders and market to lead the way.

In contrast, functional feeds sold in Europe goes through a lot of technical support and proper documentation. For example, a feed company may propose a functional feed to a farm to resolve a problem provided that the farm carries out some management changes. With educational efforts, feed companies stand to make a premium on such feeds and are liable for the performance of the feed. There is absolute transparency between feedmillers and farms and European regulations require details on the additives to be included on the label.

On developing KPIs, an independent evaluation or several independent teams (global or regional), would be the way to go. A regulatory body was also suggested. In Europe, many public-funded R&D and industry teams carry out independent studies. Similar initiatives in Asia require structuring and funding. A simpler and faster alternative is the establishment of demonstration farms and feedmills that are publicly or privately driven, but can work independently. Full transparency will instill trust between farmers and the industry. Communication is critical. With solutions, quality is extremely important but management comes first. Feeds are a tool but not the only tool farms have to control threats and conditions at the farm level.

Production Technology

The objective of nursery systems is to have better control of the younger animals during the first part of the culture cycle. These involve rearing post larvae (PL) from PL10 to PL20. Some farms have nurseries within the farm itself. After this, pre-growing continues from PL 20 to 1g juveniles over 4 to 5 weeks. Techniques differ: there are semi biofloc with very little water exchange or clear water with high water exchange. The grow-out in ponds cycle is shortened and the farm can have more crops, as there is a one-month saving for each cycle. The main weakness is the transfer from nursery to grow-out ponds. During this phase, staff require more hatchery skills than grow-out skills. Nurseries will require investments into specific infrastructure.

Education of the farmer and worker is key. Scientific knowledge is essential too. Mortality during transfer must be lower than 5%. Aside from transfer of larger and disease-free post larvae, the KPI is 80% - 90% survival rates at the end of the nursery stage. Training to manage a nursery takes time and starting with an intensive nursery system is not advisable. In determining stocking density, the knowledge and skills of workers should be considered. Should the nursery be part of a hatchery or part of the production pond? The water quality in the nursery is similar to that of a hatchery whereas pond water is usually of lower salinity

in a pond. One success cited was a Brazilian farm where PL6-PL8 were fed high protein diets to PL20 in an extension of hatchery.

Biofloc and probiotics culture systems could be complementary as they go well together and are useful for “fighting” vibrio peaks. Biofloc systems are newer techniques for intensive systems with low water exchange for stable water quality parameters. Most open systems actually have semi biofloc as with sunlight, there is competition between algae and bacteria. Biofloc systems have better waste management. Probiotics can be used in biofloc systems to establish the biofloc faster. Biofloc is good for broodstock culture as there is better control of the water environment. Broodstock gives better quality nauplii. The weakness of biofloc systems is the time required to reach stable conditions. Managing biofloc requires skills and knowledge. From the environmental aspect, pollution is low but there is a higher use of energy for aeration.

KPIs must include improvement on the stability of water parameters through routine water analysis, higher productivity, lower FCR, and less direct impact on the environment. Cost of production is another consideration.

On environment and shrimp farming, the weakness is poor collaboration between neighbouring farms. Two-way sharing of information and transparent collaboration between the farms are required. On hyper and intensive systems, the main question is the definition of intensive which vary from region to region. Intensive systems require good waste management and biosecurity. Higher productivity should go together with efficient land use. Commercial profitability should be high.

The discussion on alternative systems centered on experiences with the culture of fish with shrimp for disease mitigation. These include the farming of tilapia and shrimp. The farming with biofloc water from tilapia ponds circulated between shrimp and tilapia ponds in Brazil was discussed. The environmental issue is important for large farms, constantly under scrutiny by environmentalists and NGOs. Many farms in Thailand are now typically using only 30% of the pond area for production. The rest of the ponds are being used to store, treat and recycle water within the system. Many have tilapia in these ponds and in canals at the farm. There is a belief that this improves shrimp survival against AHPND. In terms of EMS, laboratory tests show that tilapia has some mitigating effect with APHND bacteria.

Whether these are hyper-intensive or biofloc systems and are for disease mitigation or to optimise or maximise production, the challenge is to transplant the appropriate systems to the appropriate areas. Hyper-intensive systems cannot work everywhere and the question becomes “When can a farm recuperate the investment cost?”



TARS 2017 will focus on Finfish Aquaculture: *Strategies for Growth*. It will be held in Bali, Indonesia from 16-17 August, 2017. Updates will be available at www.tarsaquaculture.com





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