Enhanced Biosecurity Measures for Sustainable Aquaculture: shrimp hatchery operations

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Long before the COVID-19 pandemic, the Broodstock Facility and Shrimp Hatchery Complex of SEAFDEC Aquaculture Department (AQD) in Tigbauan, Iloilo, Philippines, has already been practicing the best quarantine protocols. The gold standards to ensure the production of disease-free and high-quality shrimp are being developed by AQD under the program "OPLAN *Balik Sugpo*" or Operation Plan for Black Tiger Prawn Revival. Initiated in 2017, this Program generally aims to bring back the *Penaeus monodon* industry of the Philippines and help farmers revive their hopes and venture again into shrimp culture. Under the Program, the disease prevention scheme is currently undergoing verification for responsible management of shrimp broodstock obtained from the natural environment at AQD's Broodstock Facility, and for the care of the post-larval stage at AQD's Shrimp Hatchery Complex.

In the Southeast Asian region, the shrimp aquaculture industry capitalizes on the rising demand for shrimp in domestic and international markets, as the world's human population continues to grow. This lucrative industry racks up billions of US dollars in export income annually, making it a consistent money-maker for aquaculture farmers from 2008 to 2017 (**Figure 1**). However, the industry is very delicate and fragile as the culture species solely rely on their innate immunity to protect against pathogens. The lack of adaptive immunity in shrimps (Amar & Faisan, 2012) and the emergence of new diseases or the recurrence of old ones can entirely wipe out shrimp production, which could lead to the collapse of the industry.

Cultured shrimps are vulnerable to diseases that result in physical abnormalities, stunted growth, reduced fecundity, and mortality. Several causative agents include the bacteria like *Vibrio harveyi* and *V. splendidus*, which cause bacterial and viral diseases such as the baculovirus which gives rise to the white spot syndrome virus (WSSV) disease and parvovirus that brings about the infectious hypodermal and hematopoietic necrosis virus (IHHNV) disease (Lavilla-Pitogo *et al*., 2000; de la Peña *et al*., 2015). The susceptibility of shrimps to a number of pathogens means that shrimp farms should definitely include biosafety procedures in their culture practices.

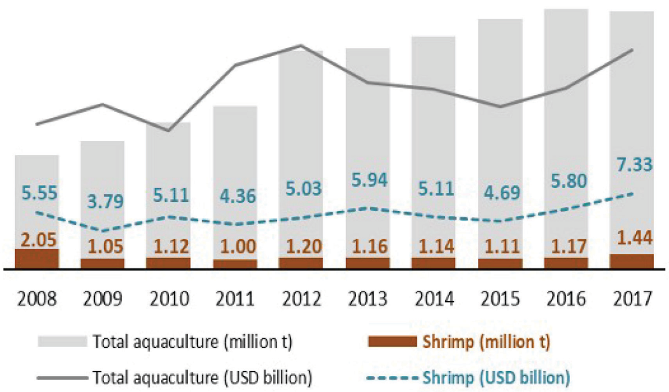

Figure 1. Shrimp culture and aquaculture production of the Southeast Asia region in 2008-2017 by quantity (million t) and value (USD billion) (excluding freshwater prawn)
Source: *Southeast Asian Fisheries Development Center (2020)*

Adding fuel to the fire, the rapid intensification of shrimp farming in recent years to produce more shrimp without regard for good aquaculture practices (GAqP) has allowed disease outbreaks to occur, ultimately translating to reduced production and financial losses. For example, the Philippines used to be at the forefront of *P. monodon* culture in the Southeast Asian region. However, the country's *P. monodon* industry quickly collapsed at its peak largely due to intensified and irresponsible farming, pollution, environmental degradation, and disease outbreaks (Golez, 2009; Rosario & Lopez, 2005).

OPLAN *Balik Sugpo*

In the past, milkfish has dominated the Philippine aquaculture scene, and shrimps and prawns were just seen as incidental crops (Gicos, 1993; Primavera, 1992). Farmers soon realized the market potential of shrimp particularly the black tiger shrimp (*P. monodon*). So, the Philippines, in the 1960s started exporting black tiger shrimps to Japan which then led to a boom in production due to the high demand of shrimps overseas (Hishamunda *et al*., 2009; Rosario & Lopez, 2005).

During the 1970s, the life cycle of *P. monodon* was first completed in captivity by SEAFDEC/AQD using eyestalk ablation to induce maturation. As time goes by, more research and development milestones have been achieved by AQD. Extension manuals on pond culture of *P. monodon* were published, as the mass production of spawners through eyestalk ablation in marine pens was achieved. The technology on mass production of fry was also developed and disseminated through regular training courses leading to the establishment of small-scale hatcheries not only in the Philippines but also in the Southeast Asian region.

In the 1980s, the production of spawners through ablation was achieved in land-based tanks giving rise to the publication of the manual on broodstock management and a technical report on the biology of the shrimp, *P. monodon*. In addition, an effective and cost-effective diet for grow-out was developed, and when the 1990s came, a microparticulate diet was formulated for the shrimp larvae, while the mangrove-friendly shrimp farming techniques were developed. In late 1990s, however, when diseases hit the shrimp hatchery hard due to intensification of stocks and irresponsible culture practices in ponds, there was a huge decline in the country's *P. monodon* that devastated the Philippine shrimp industry affecting the shrimp growers and making the hatchery operators cease their fry production.

At present, AQD is embarking on several activities to revive the shrimp industry of the Philippines under the program "OPLAN *Balik Sugpo*" which aims to produce high-quality *P. monodon* postlarvae (PL) to supply the shrimp farmers for grow-out culture. AQD has enhanced the existing culture procedures particularly applying strict biosecurity measures and instilling GAqP in its shrimp hatchery operations. Under the Program, the current disease prevention scheme (**Figure 2**) includes verification for the management of the shrimp broodstock obtained from the natural environment at AQD's Broodstock Facility, and the resulting PL for rearing at AQD's Shrimp Hatchery Complex.

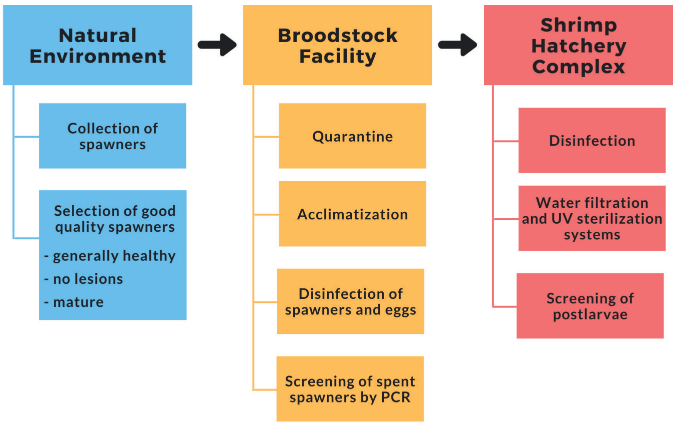

Figure 2. Disease prevention scheme of the program "OPLAN *Balik Sugpo*" of SEAFDEC/AQD

Natural Environment

Broodstock (spawner) of shrimps are usually acquired from the wild by means of trawling between 12 m and 55 m water depth. The good spawners are selected, which should be generally healthy, have no cuts or damages, and most importantly, mature enough to spawn overnight (*i.e.* each gravid female shrimp holding 200,000–1,000,000 eggs). Then, the selected spawners are transported in fry bags supplied with aerated seawater to AQD's Broodstock Facility.

Broodstock Facility of AQD

The Broodstock Facility of AQD for shrimps is located at the Tigbauan Main Station of SEAFDEC Aquaculture Department in Iloilo, Philippines.

Disinfection of staff and guests

At AQD's Broodstock Facility, the staff must wear personal protective equipment (PPE) including face masks, gloves, scrub suit, laboratory gown, and boots (**Figure 3**). The doors of the Broodstock Facility are also installed with alcohol dispensers and foot baths. The staff as well as guests may be carrying with them pathogens from the outside so they should also frequently sanitize to minimize the exposure of the shrimp spawners to pathogens.

Figure 3. A staff at AQD's Broodstock Facility in complete personal protective equipment

Disinfection of spawners and eggs

Upon arrival, the spawners are housed at the Broodstock Facility which is located in a building separate from the Shrimp Hatchery Complex to avoid cross-contamination of pathogens. The spawners acquired from the wild are possible carriers of diseases, thus, they are subjected to strict quarantine procedures at the Broodstock Facility to make sure that the contamination of pathogens is controlled.

For acclimatization, the spawners are placed in white basins filled with aerated water adjusted to equalize with the salinity of the water in the transport bag. When both of the water salinities have equalized, the spawners are allowed to remain in the basin for two hours. After acclimatization, the spawners are disinfected for 15 min with 50 ppm formalin or povidone-iodine added to the water in the basin (**Figure 4**). Then, after

Figure 4. A hatchery staff adding formalin to disinfect the spawners

checking for gonadal maturity, one spawner is placed in each fiberglass spawning tank and allowed to spawn overnight. Then the individual spawning tanks are covered with black nets and black sacks to avoid entry of other organisms and contamination between the tanks.

Each spawning tank is equipped with a set of materials including dipper, scoop net, harvesting box, and others to be used during the spawning operations. After spawning, the eggs are washed, this has long been practiced, to reduce the accumulation of bacteria and viruses when the eggs hatch into nauplii (Licop, 1988). It is said that the capsule that encloses an egg might contain bacteria and viruses during spawning. During hatching, the capsule breaks and the eggs turn into nauplii. If the eggs are not washed and disinfected, the nauplii may engulf the bacteria and viruses as their mouths begin to open. In the current practice, egg washing procedure has been enhanced to make sure that the eggs are free from pathogens before hatching into nauplii. The eggs are washed with running UV-sterilized seawater for five minutes in the first basin, and then transferred to a second basin with aerated seawater added with 50 ppm of povidone-iodine solution. Finally, the eggs are placed in a third basin and rinsed again with running UV-sterilized seawater for five minutes to eliminate the pathogens that could have attached on the surface of the eggs.

Screening of spent spawners and eggs

In earlier times, disease detection in shrimp farms was difficult to carry out, and in most cases it was already too late when diseases are detected. Nowadays, disease surveillance has been improved with the advent of polymerase chain reaction (PCR). The tissue samples of spent spawners and egg samples are sent to the Laboratory for pathogen detection through reverse transcription polymerase chain reaction (RT-PCR) technique to determine the presence of pathogens and diseases, such as WSSV, IHHNV, EHP, AHPND, monodon baculovirus (MBV), yellow head virus (YHV), and others.

AQD Shrimp Hatchery Complex

At the Shrimp Hatchery Complex of AQD (**Figure 5**), the biosecurity practices and facilities had been enhanced and modified to keep up with the increasing demand for shrimp fry, prevent the occurrence of diseases, mitigate the potentially harmful damage caused by the pathogens, and reduce mortality rates.

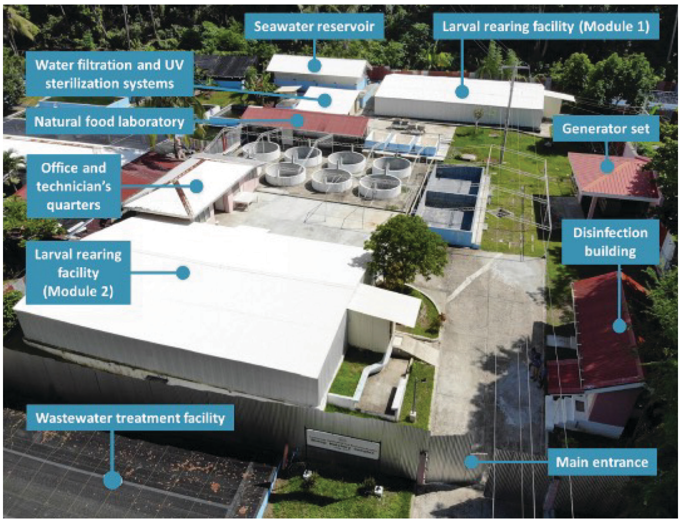

Figure 5. Shrimp Hatchery Complex of SEAFDEC/AQD at the Tigbauan Main Station, Iloilo, Philippines

Disinfection of staff and guests

Aside from isolation and routine testing of spawners, eggs, and fry, biosecurity measures in hatcheries are now focused also on the staff and guests who should be responsible in minimizing the exposure of fry to pathogens. Before entering the facilities at AQD's Shrimp Hatchery Complex, the staff and guests are required to take a shower at the disinfection building and wear PPE. Alcohol dispensers and foot baths are also installed near the doors of the hatchery facilities for disinfecting hands and shoes, as this could help eliminate pathogens that may have been carried by the staff and guests from outside.

Water filtration and UV sterilization systems

The hatchery is utilizing additional technologies to make sure that microscopic pathogens like bacteria and viruses could not reach and affect the stocks. The seawater pipe and aeration systems have been improved and equipped with ultraviolet (UV) light sterilization system. As shown in **Figure 6**, the pumped seawater would have to undergo several filtration systems before reaching the larval rearing tanks. First, the pumped water from the sea passes through the sand filter before reaching the 180-ton concrete reservoir. Then, the seawater from the reservoir passes through the UV sterilizer to make sure that pathogens are eliminated or become limited

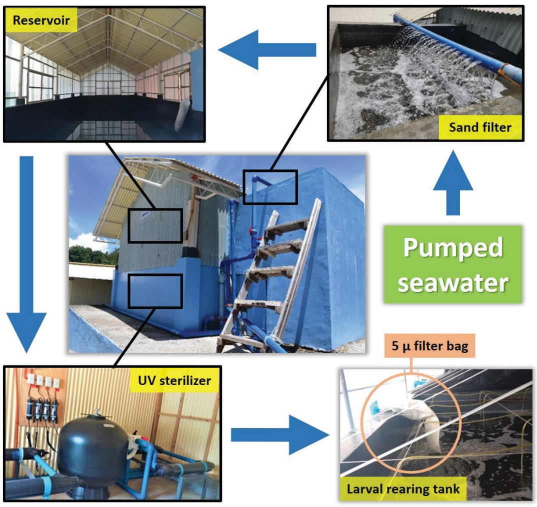

Figure 6. Water filtration and UV sterilization systems at the Shrimp Hatchery Complex of SEAFDEC/AQD

before reaching the larval rearing tanks which are installed with 5 µm filter bag. Finally, the sterilized water is used for rearing the stocks and culturing the natural food for the larvae.

Screening of postlarvae

Using clean and covered pails, all nauplii are harvested and transported from the Broodstock Facility to the Shrimp Hatchery Complex in an order that takes into account the results of the PCR analysis. First, all the nauplii of spawners with negative PCR test results are harvested and stocked in Module 2 of the larval rearing facility. Subsequently, the nauplii of pathogen-positive spawners are harvested and stocked in Module 1 of the larval rearing facility. All the larval rearing tanks are covered with black sacks to avoid entry of other organisms and avoid contamination between the tanks. Cleaning and disinfection of facilities and equipment are also performed to remove all pathogens present in a certain area that might affect the cultured shrimp. All stocked shrimp larvae are reared and treated equally. Particular staff is assigned to feed and monitor the stocks in each module. Daily monitoring of larvae is carried out to detect any problems and diseases that might occur. Samples from the stocked nauplii and the rearing water are subjected to bacterial analyses twice a week for surveillance to detect the presence of luminous bacteria, other *Vibrio* species, and other pathogens.

To ensure that no pathogen escapes detection, the fry are subjected to routine RT-PCR testing for WSSV, MBV, IHHNV, YHV, APHND, and EHP when they reach PL 5, PL 10, and PL 15. This protocol also ensures that none of the fry are potential disease carriers when sold to aquafarmers for grow-out culture.

If results come back positive, the fry is harvested and stocked in Module 1. This practice is deemed sustainable because none of the pathogen-positive nauplii are discarded, but instead, reared and monitored to become high quality and disease-free fry (**Figure 7**).

Figure 7. High quality and disease-free *Penaeus monodon* fry produced at the Shrimp Hatchery Complex of SEAFDEC/AQD

Way Forward

During 2019–2020, SEAFDEC/AQD produced around 3.06 million high quality and disease-free *P. monodon* fry. Relating to the OPLAN *Balik Sugpo*, the disease-free fry produced from the hatchery had been stocked in ponds at Dumangas Brackishwater Station of AQD for the verification run using environment-friendly culture techniques. This yielded about 15 t of black tiger shrimp, which was a step forward to reviving the black tiger shrimp industry starting in Iloilo, Philippines, and hopefully to the whole country and throughout the Southeast Asian region in the future.

The OPLAN *Balik Sugpo* program also focuses on other important factors such as feeds and disease treatments for the successful revival of the black tiger shrimp industry in the country. In the 5-year road map (**Figure 8**) presented by SEAFDEC/AQD during the Shrimp Road Map Updating Workshop conducted by the Philippine Bureau of Fisheries and Aquatic Resources, the program "OPLAN *Balik Sugpo*" would continue with the verification of the improved hatchery and grow-out technologies at the local level and eventually at the national level.

SEAFDEC/AQD has also started with the development of cost-effective feeds for shrimps using locally sourced materials and would proceed with its field verification by the end of 2021. Meanwhile, a vaccine for WSSV is being developed but its development is still at the R&D stage. By 2024, all of the endeavors under the Program should hopefully be ready for rollout. Then, the SEAFDEC Member Countries could request for the transfer of the technology through training and other relevant information activities, through the SEAFDEC mechanisms.

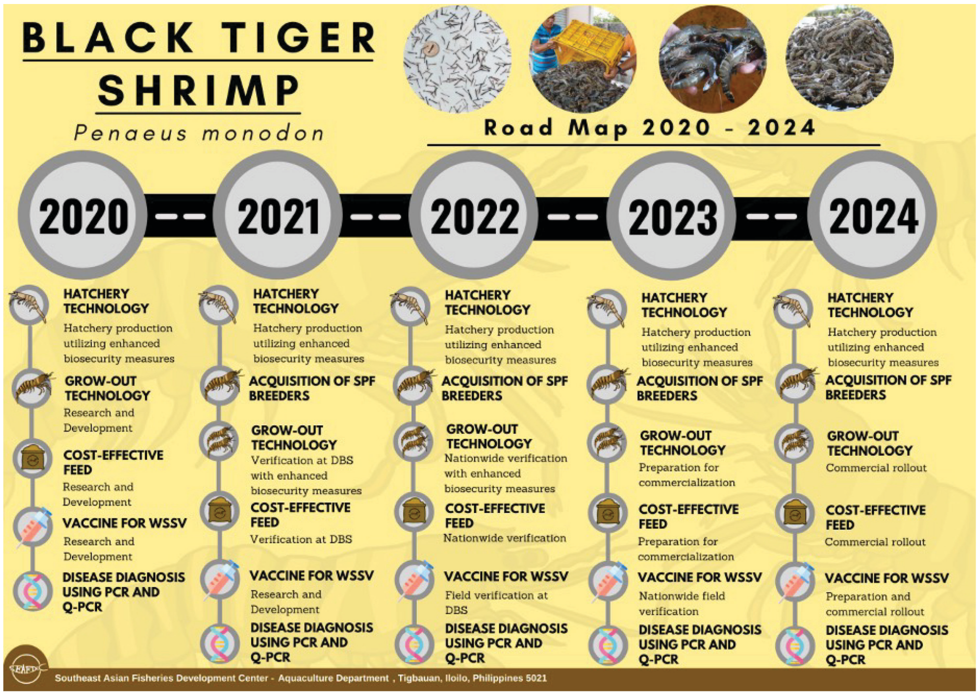

Figure 8. SEAFDEC/AQD 5-year road map for black tiger shrimp under the program "OPLAN *Balik Sugpo*"

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