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Genetic Research Initiatives for Sustainable Aquaculture Production in the Philippines

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Abstract

The Philippines is one of several countries in Southeast Asia that has, for several decades, made steady contributions to world aquaculture production both from inland and marine waters. In recent years, fish production has been on the decline mainly because of the lack of quality seedstock, limited stocks of captive breeders or spawners of major aquaculture commodities, adverse effects of climate change and other environmental factors on fish breeding and rearing, fish diseases caused by pathogenic organisms and prohibitive cost of aquaculture inputs such as feeds, etc. Genetic researches have been conducted mostly through local grants with the aim of addressing the aforementioned constraints. Such initiatives focused on developing and applying methods in (a) selective breeding; (b) marker-assisted genetic strain assessment for broodstock development and for monitoring inbreeding in farmed stocks and (c) genomics to understand and enhance on-farm stock performance through the identification of genes that are responsible for nutrition, stress and immune responses, among others. This paper highlights examples of local genetics applications in tilapia, mangrove crab, shrimp, milkfish and abalone aquaculture. The significance of implementing genetic interventions to boost and sustain aquaculture production in the Philippines is likewise discussed.

1. Introduction

In the past decade, aquaculture production has gradually increased to levels that represent almost 50% of the total global fish production (FAO, 2018). Of the total world aquaculture production, 89.2% or 71.5 million tonnes come from Asia, with Philippines ranking as the 7th major contributor, having 796,000 tonnes produced in 2016. This, in spite of the challenges that the aquaculture industry has faced, is currently experiencing. FAO (2018) has projected a growth in Philippine aquaculture production of 36,300 tonnes by the year 2030. This can be achieved if the industry finds solutions to problems pressing both aquaculture and fisheries, such as poor quality seedstock and broodstock, costly and/or inferior diets, poor husbandry methods, among others. Recognizing the problems besetting aquaculture, local research and academic institutions have continuously worked on either environmental and/or genetic interventions to ensure sustainable aquaculture.

2. Environmental interventions to increase aquaculture production

Farmed fish production involves four phases, namely: (a) broodstock development, management and breeding, (b) hatchery seed production, (c) nursery rearing, and (d) grow-out culture. Each phase requires environmental interventions to optimize fish yield. Knowledge on proper nutrition and maintaining optimal water quality conditions are essential from broodstock development and management to grow-out. However, the major phases that can benefit from appropriate environmental interventions are at the nursery and grow-out phases since these are when one needs skills in good aquaculture practices and/or husbandry such as applying the correct fish stocking rate, feeding schemes, water quality and fish health management etc.

3. Genetic solutions for improved fish production

Genetic technologies have for some time, provided options for aquaculturists to improve their fish farm output. The Philippines have embarked on several projects, some internationally funded, to boost local fish production by way of genetic programs. Such projects focused on major aquaculture species such as tilapia, milkfish, shrimp and mangrove crabs. Apart from these, species that may not be widely farmed but are important (e.g. abalones) have been subjects of genetic studies in view of the need for their populations to be genetically characterized and propagated for resource conservation and enhancement. Some of the genetic methods include selective breeding, DNA marker assisted broodstock development and management and the more advanced technologies such as genomics. Applications of these methods shall be discussed in the foregoing sections, that is, according to species.

4. Increasing tilapia production through genetic strain improvement

Tilapias, particularly of the genus *Oreochromis,* have been introduced into Asia from Africa as early as in the 1950s, starting with the highly prolific *Oreochromis mossambicus*. Three species, namely *O. mossambicus*, *O niloticus* and the red tilapia hybrids (*Oreochromis* spp) are important in Philippine aquaculture however of the three, the Nile tilapia (*O. niloticus*) has been farmed widely, not minding if the species is not indigenous as it has been cultured in different enclosures (tanks, lake-based cages, and ponds) and are now found in freshwater lakes and other inland waters. The popularity of the species peaked in the early to mid-1980s when the Philippines even became the highest tilapia producing country in the world. Gradually, tilapia farm yields declined and the reduction has been attributed to factors that primarily include genetically depauperate (e.g. inbred) stocks that have become slow-growing and less fit. This has prompted the government fisheries agency, public as well as private academic and research institutions to work together to develop genetically enhanced tilapia stocks to support the need of the tilapia farms for quality, fast growing seedstock (SEAFDEC, 2017). **Table 1** lists some of the tilapia strains (GIFT, GMT, FaST) that have been developed in the Philippines (SEAFDEC, 2017; Asian Development Bank, 2005; WorldFish Center, 2004; Mair *et al*., 1995). The improved strains have been disseminated locally and have also been exported to other tilapia producing countries after private entities (GIFT Foundation International Incorporated, Genomar, PhilFishGen) took over the operations from the agencies that developed them. In 2004, several of the improved Nile tilapia strains and red tilapia hybrids have been genetically characterized using mtDNA-RFLP and microsatellite markers (Romana-Eguia *et al*., 2004). Results have been used to confirm which among the strains have high genetic diversity and such information have been used to validate the good performance the strains have in culture. Information as well on the use of genetic markers to monitor inbreeding in selected stocks have been generated (Romana-Eguia *et al*., 2005). This has generally been used to demonstrate evidence of increased inbreeding rate in subsequent generations of a mass selected tilapia strain developed from a limited founder stock. Such studies could have practical implications on how selected fish in general should be managed especially when kept and used as hatchery broodstock.

Overall, the tilapia genetic improvement programs have been beneficial to the tilapia industry in that during the period that several selected breeds were disseminated, tilapia production gradually increased not only in the Philippines but also in Bangladesh, China etc. where some of them were made available (FAO, 2018).

5. Marker-aided broodstock management to improve milkfish production

Milkfish is a commercially important commodity in the Taiwan, Indonesia and in the Philippines (where it is

Table 1. List of genetically improved strains of Nile tilapia (*Oreochromis niloticus*) developed in the Philippines (modified from SEAFDEC, 2017).

STRAIN	GENETIC PROGRAM/METHOD
GIFT Tilapia	Genetically improved farm tilapia (GIFT) program: Combined family and within family selection for improved growth
Genomar Supreme Tilapia (GIFT-derived)	Genomar Project: Combined selection for improved growth, marker assisted selection
GET Excel and **iExCEL** or improved GET Excel stocks	GET-Excel Program:Outcrossing two fast-growing strains (FAST and GIFT) for improved growth
Genetically Male Tilapia or YY supermale tilapia	Genetically Male Tilapia (GMT) Program: selective breeding and sex reversal methods
Brackishwater Enhanced Selected Tilapia (BEST) and i-BEST or improved BEST	Brackishwater Enhanced Selected Tilapia Program: hybridization and outcrossing; size-specific selection for salinity tolerance
Cold tolerant tilapia	Cold tolerant tilapia: hybridization
FAST Tilapia	Freshwater Aquaculture Center Selected Tilapia Program: rotational mating and hybridization
SaltUno tilapia straina	Molobicus or SaltUno project: hybridization to produce salt tolerant tilapia

considered as a national fish). In the Philippines, successful completion of the life cycle in captive milkfish stocks was achieved in 1975 and several generations have been produced from thence owing to research breakthroughs at the Aquaculture Department of the Southeast Asian Fisheries Development Center (SEAFDEC/AQD) where milkfish R&D work continues to date. Earlier studies focused on milkfish reproductive physiology and later, on the development of hatchery, nursery and grow-out methods. At one point, the government attempted to harmonize efforts in milkfish seedstock production among public hatcheries by establishing a National *Bangus* (local term for milkfish) Breeding Program. This initiative met some funding problems well into a decade and a half from the time the program was established that prompted the government hatcheries to sell their breeders to private farms. At present, SEAFDEC/AQD has broodstock that are close to four decades old and are still spawning. Several private milkfish farms either procure stocks for culture and/or propagation from Indonesia, which has a well-organized system of fry production that enables them to sell their fry at a cost lower than Philippine sources. It has been noted that in the past few years, the Philippines has been importing more than 50% of its two billion annual fry requirements from Indonesia. This has therefore encouraged some work on the DNA marker-based genetic assessment of wild and hatchery stocks to enable local farmers to identify good sources of either broodstock. material and/or seedstock for on-growing to marketable sizes, in their farms. A study on milkfish genetic variation showed that the local milkfish breeders (both from the wild and from the hatchery) except those kept as mixed generation stock in a government hatchery facility, have genetic diversity levels comparable to the imported Indonesian stock (Romana-Eguia *et al*., 2018). It is indicative therefore of the fact that since the imported stock is initially perceived to be better than our own, the Philippine farmers can very well rely on the domesticated stocks for providing fry/fingerlings to address the increasing industry demand for good quality milkfish seedstock.

6. Genetic and genomic applications in Mangrove crab farming

Mangrove crab aquaculture in the Philippines is a growing industry although mangrove crab production ranks 9th among the commonly farmed aquatic species in terms of quantity. Nonetheless, the local and export market demand for this commodity (either live and hard-shelled or frozen and soft-shelled forms) has been increasing thus warranting particular attention in terms of improving its current supply. The mangrove crab species found in the Philippines are three, namely *Scylla serrata* (a fast growing hence most preferred species), *S. tranquebarica* and *S. olivacea*. *S. serrata* has been well-studied; from domestication, genetic stock characterization (Quinitio *et al.*, 2011) and recently, selective breeding. These notwithstanding, more studies are still being conducted and these involve basic researches that are geared towards the development of technologies that have practical applications. For example, juvenile stages of the three local species are difficult to distinguish from each other. Hence, a mobile phone application, known as "Crabifier" was developed for crab collectors, traders and crab farmers. This is based on molecular marker data and image analysis (Vince Cruz-Abeledo *et al*., 2018). Other genetic/genomic tools that have been applied in mangrove crab farming research have generated valuable data described in what the proponents conveniently refer to as "CrabMAP", "CrabADAPT", "CrabMOLT" and "CrabSNP" (DOST-PCAARRD, in press). Briefly, "CrabMAP" refers to temperature vulnerability maps for local key areas where mangrove crabs are farmed. Such information will be valuable in developing climate smart aquaculture, in this case for rearing *Scylla* crabs. On the other hand, "CrabADAPT" refers to information on genetic adaptability of certain mangrove crab populations when exposed to wider temperature range and anomalies. The idea is based wholly on gene expression patterns across populations that are exposed to different temperature profiles. Meanwhile, "CrabMOLT" refers to the study where optimal salinity and temperature combinations that would favor frequent molting in crabs were investigated. Frequent molting would be indicative of crabs that could be fast growing. It is in this study where the ratios of a molt-promoting hormone (ERK) to a molt-inhibiting hormone (MIH) were established for the different molting stages of the mangrove crabs. Finally, "CrabSNP" is a study on the identification of single nucleotide protein (SNP) variants associated with adult sized immature female crabs, and identified genome markers that may be associated with such phenotypes. Immature female mangrove crabs command a high price in the local trade. All these novel genetics based technologies shall be further validated with more farmers the aim of helping them improve their production.

7. Genetic tools in shrimp health management

The tiger shrimp *Penaeus monodon* has for some time been a major export income earner for the Philippines prior to the industry experiencing production slumps due to shrimp viral diseases e.g. AHPND or acute hepatopancreatic necrosis disease (De La Peña *et al*., 2015). Several large-scale commercial shrimp producers have access to imported specific pathogen free (SPF) shrimp from e.g. Hawaii, however small to medium scale shrimp growers have no recourse but to procure wild and/or hatchery-bred seedstocks with the risk of using juveniles that are highly susceptible to diseases. In this regard, local shrimp culturists are advised to report pathogens found in Penaeid shrimps in their farms through a website referred to as Online Philippine Shrimp Pathogen Information Resource (OPSPIR; opspir.seafdec.org.ph) developed by UP, Ateneo de Manila University, the Philippine Genome Center, the Bureau of Fisheries and Aquatic Resources, the Department of Science and Technology and SEAFDEC/AQD. This initiative is a countrywide bio-surveillance system that will enable the documentation and mapping of areas where shrimp farms are affected by specific pathogens. Once reported, validation of the reported occurrence can be made on samples collected on-site by using PCR-based diagnostic tools while referring to genome sequences in the OPSPIR database.

8. Genetics for abalone aquaculture and stock enhancement

The abalone or "awabi" in Japanese is a highly-priced marine mollusk. In the Philippines, SEAFDEC/AQD has, since 1994, studied the indigenous species known as donkey's ear abalone or *Haliotis asinina*. Although culture techniques and requirements have already been established locally, the industry has yet to achieve steady growth. The lack of information on good sources of quality abalone seedstock and broodstock could somehow explain its limited production from aquaculture. A study aimed to generate a preliminary database on possible sources of genetically diverse *Haliotis asinina* stock was conducted. Samples from one hatchery-bred and nine wild sourced founder stocks and their F_1 offsprings were analysed for genetic variability using microsatellite markers. Together with growth data from the same, information from this study shall be used to identify local stocks most suitable for breeding and farming.

9. Conclusion

It is with much optimism that the Philippine aquaculture industry views the foregoing genetic initiatives to boost fish production. The impact created by the adoption of genetic improvement schemes on tilapia production in recent years could prove the advantage/benefits of using such interventions. Aside from conventional selective breeding methods, recent developments and tools in genetics research such as genomics, gene editing etc. as applied in aquaculture could hasten and sustain its growth and ultimately address the global need for food security.

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