Milkfish and Tilapia as Biofactories: Potentials and Opportunities

Mudjekeewis Santos, Irma Destura and *June Feliciano Ordoñez*

Living organisms used in commercial-scale production of compounds such as recombinant proteins for agricultural, biomedical, and pharmaceutical applications are referred to as biofactories. This article assesses the role of two most important aquaculture species in the Philippines, *i.e.* milkfish and tilapia, as prospective biofactories from their production to semi-processing chain, considering the economic importance of these resources in the country. Based on a paper presented by the authors during the Round Table Discussion on Marine/Aquatic Biofactories in the Philippines organized by the Philippine National Academy of Science and Technology on 13 March 2013, this article focuses on the status of production and utilization of these species, especially on how these are utilized and processed into value-added products, as well as points out the underlying issues and concerns that impede the sustained role of these species as biofactories.

The rise of human population parallels with the growing need of the essential necessities of everyday life, which might not be limited only to food, shelter, and clothing, but also novel products that are made available through the integration of science and new technology-based systems. In our society today, many products are produced using raw materials generally coming from and already existing in the natural resources which are known to be free but their quantities may be continually declining. In order to maintain and enhance the availability of such resources, their production is always being intensified while their availability as raw materials for creating novel products should be assured. Two most economically-important fisheries commodities in the Philippines, *i.e.* milkfish and tilapia, have been found to have the potentials as biofactories.

Plant, algal, and bacterial cells have been the most successful biofactories, and are utilized in the production of many important metabolites in both wild type form or as recombinant cells (Sarmidi and El Enshasy, 2012). Sweeteners, essential oils, agar, carrageenan, biodiesel, antibiotics, and recombinant proteins are among the compounds produced from various biofactories and are now used for different purposes in various industries. Interestingly, several types of commercially-valuable compounds can also be extracted from fish especially from fish processing by-products using biotechnology. This opportunity could drive the Philippine tilapia and milkfish industry from being limited to fish meat production, as these aquatic species had been considered for decades, towards becoming biofactories. Certainly, the future progress of both industries will not only come from increased production volume and current value-added products but also by generating alternative products which are mainly offered through biotechnology techniques.

Current Milkfish and Tilapia Industry Statistics in the Philippines

Milkfish

Although not fully determined, the earliest account of the development of milkfish industry in the Philippines pointed to its existence even before the arrival of the European colonizers in the 1500s. The early milkfish farming then was more of a trap-and-grow operation based on the natural stock of milkfish fry that comes inland with the tidal waters. In 1900s, milkfish farming was purely a private sector effort in many areas of the country, namely: Central Luzon, Pangasinan and Iloilo Provinces. Milkfish culture from the early days of American rule (Radcliffe, 1912; Day 1915; Herre and Mendoza, 1929) until post independence in 1946 was mainly described based on such existing traditional practices.

The reorganization of the former Bureau of Fisheries in 1947, led to the conduct of research on milkfish culture focusing on fertilization and *lablab* production (Rabanal, 1949). In 1968, a hatchery project in Naujan, Mindoro was developed in order to minimize total dependence on natural supply of fry. In the early 1970s, milkfish culture in pens began in Laguna Lake (Delmendo and Gedney, 1974), and was found to be successful and commercially viable. In late 1970s, the Philippine Bureau of Fisheries and Aquatic Resources (BFAR) under a UNDP-funded project developed a production calendar to guide milkfish farmers in different climatic zones of the country. In 1981, the National Bangus Breeding Program (NBBP) by BFAR and SEAFDEC was established to jumpstart the mass production of milkfish fry and demonstrate its technical and commercial viability.

Status and trend of milkfish production in the Philippines

About 98% of milkfish production in the Philippines comes from aquaculture with only a very small amount from capture fisheries. Production from milkfish culture continued to increase contributing about 15% to the total aquaculture fish production of the country. An increase of about 4% production in 2012 from 2011 was a result of milkfish good farm management, availability of quality fry/fingerlings, and proper feeding practices (BAS, 2012). From 2002 to 2011, production of milkfish in aquaculture (**Fig. 1**) grew at an average rate of about 3% (PCAARRD, 2012), where production from brackishwater fishponds was known to be the highest among the various production systems.

The country's top five milkfish producing provinces are Pangasinan, Capiz, Iloilo, Negros Occidental, and Bulacan (**Fig. 2**). While Pangasinan had the biggest share of the production at 39%, Capiz, Iloilo and Negros Occidental come next contributing about 30%, and then Bulacan accounting for about 11%. In 2010, milkfish export amounted to 4,626 MT valued at PHP 715.05 million, where 60% of total exports were in frozen form and 25% were in whole or in pieces, and fillet and frozen forms in minimal quantities (BAS, 2012). A number of private and government milkfish hatcheries are operating in the Philippines, but despite their existence, some farmers continue to import milkfish fry from Indonesia and Taiwan. (PCAARRD, 2012). Recently, there has been an increasing trend in the utilization of milkfish in the country because of its availability in local markets.

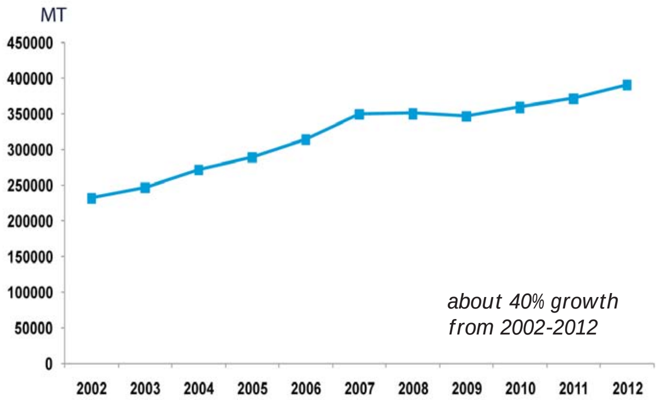

Fig. 1. Total annual milkfish production of the Philippines from 2002-2012 (BAS, 2012)

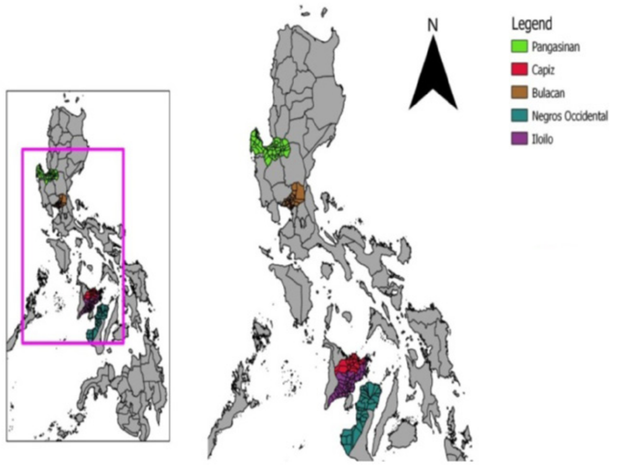

Fig. 2. Five top milkfish producing provinces of the Philippines

The performance of the milkfish industry has been affected by the limited supply of quality fry, especially considering the notable decline in the fry supply from the wild (Ahmed *et al*., 2001; Bagarinao, 1999) recorded during the previous years. Nevertheless, the demand for fry has been growing due to culture intensification and shift in production towards milkfish farming in reaction to the dwindling prawn industry (Israel, 2000). According to Israel (2000), if milkfish production is to keep pace with the average annual national population growth of about 2.5%, the country will have to produce about 356 million more fry in 2005 and 617 million more in 2010. The required volumes of fry will increase if there would be a decline in the available wild fry from the 1997 level or if other objectives beyond just meeting the needs of the growing population are targeted as well. To achieve this, there is a need to: (a) seed the open water bodies, (b) lower the nutritional deficiency rates of the population, and (c) enhance exports.

Israel (2000) also noted that if the growth rate in milkfish production is aimed at 5% annually, the additional fry requirement will go up to 1,443 million by 2010. Nonetheless, he also offered options to address the problem of limited supply of milkfish fry, *i.e.* either through importation or development of a home grown industry that will produce hatchery-bred fry in sufficient quantity and quality. While he considered the first option as undesirable since it can lead to transporting into the country certain milkfish diseases that are not locally endemic and will also cost the country its much-needed foreign exchange, it will forfeit the chance of exploiting the country's natural comparative advantage in aquaculture.

The development of an industry that produces hatchery-bred milkfish fry could address the problem of limited fry supply over the long term, and also helps in avoiding the undesirable effects of importation as well as decrease the price of fry and milkfish products in the long run. The effect of these two options will be essential since it can promote the competitiveness of local milkfish-based products in the domestic and international markets.

The increasing demand for milkfish fry in mid 1990s when fish pens and cages in brackish and marine waters started to appear had nevertheless, prompted investigations into artificial spawning of milkfish broodstock in captivity. Therefore, based on the results of the Bangus Fry Resource Assessment in the Philippines conducted in 1996-1997, research studies have been conducted from 2000 up to the present, to increase the volume of fry from local hatcheries, and improve fry quality and performance to make the country's milkfish industry competitive.

Utilization of milkfish in the Philippines: value-added products

Value-adding is defined as increasing the worth or value of a product after it has undergone simple or complex processing operation, and turning simple products into value-added products in order to obtain better income, improve processing utilization and provide variety of products keeping at pace with consumers' needs (Alsons Aqua Technologies Inc., 2004). In milkfish, the most common form of value-adding occurs in filleting, deboning, smoking, and marinating the fish, the products of which are packed and sold chilled or frozen (Yap *et al*., 2007). Nowadays, filleted, deboned and smoked milkfish products are sold not only in local but also in international markets, especially in the USA, Japan and other neighboring Asian countries. Some processed products of milkfish are also exported to other European countries (Alsons Aqua Technologies Inc., 2004). The deboned form, locally known as "*boneless bangus*", is the most popular among the value-added products of milkfish. The by-product of the boneless bangus such as trimmings and bits of flesh that are removed with the bones, are combined to pay forward to another forms of local processed products such as fishballs, milkfish *lumpia*, *quekiam*, and *embutido*, while the milkfish skins are turned into *chicharon* (Yap *et al*., 2007).

Nowadays, commercial companies engage in canning industry are developing new forms of processed products using milkfish as main material, which are also gaining popularity because of the availability of raw materials, the milkfish which is grown locally in the country. The new processed canned milkfish product which is already available in the local market comes in different flavors derived from famous Filipino foods. The added ingredients are mixed with raw milkfish, and undergo cooking and several stages of processing techniques to make the final product more tasty and palatable. In some provinces, like in Pangasinan, not only is the milkfish "meat" utilized for value-adding but also the internal organs (intestines, lungs, heart, and stomach) which are used to produce "*bagoong*" (fish sauce) or fish paste, a famous condiment in Filipino cuisine. In local pastry shops, cookies mixed with milkfish bone have been developed and sold as calcium-rich snacks for kids while they are starting to develop their bones, and for adults needing additional calcium supply.

Tilapia

Tilapia culture in the Philippines began with the introduction of Mozambique tilapia (*Oreochromis mossambicus* Peters 1852) imported from Thailand in 1950s. However, culture of this "wonder fish" as it was called back then, failed to expand in the commercial production because of its unwanted characteristics such as early maturity resulting to overpopulation in fish ponds, stunted growth, small in size at harvest, became "pest" in brackishwater ponds, and unappealing dark-color (Bolivar, 1993; Guerrero, 1994). Therefore, the country's production of tilapia in 1960s was minimal (FAO, 2006), undermining the slow progress of tilapia farming that was not revived until a decade later (Yosef, 2009).

In 1970s, the fuel oil crisis severely damaged the country's marine fisheries industry (Guerrero, 1994), forcing the Philippine Government to give a higher priority in raising fish production from inland aquaculture to cover for the impending shortage of fish products. Tilapia was then chosen for such development because of its many desirable characteristics compared to other aquaculture fishes and its potential to benefit the resource-poor rural people as well as commercial growers. This is considering also that since 1972, different strains of Nile tilapia (*Oreochromis niloticus* L.) had been introduced to the country (Guerrero and Tayamen, 1988; Bolivar, 1993). Nile tilapia therefore, rapidly gained popularity to farmers and consumers because of its better characteristics (*e.g.* lighter color, faster growth, and high tolerance to various environmental conditions) over the Mozambique tilapia.

During that time, many developing countries were confronted with major constraints in tilapia culture that include inadequate supply of seeds and lower genetic quality of cultured stocks compared to the wild population because of inbreeding depression (Pullin and Capili, 1988; Eknath *et al*., 1993; Acosta *et al*., 2006). Thus, tilapia production during the 1980s continued to decline due to deterioration of the genetic quality of stocks that led to the significantly reduced performance of farmed Nile tilapia. Meanwhile, the public sector, national institutions, and international organizations based in the Philippines initiated selective breeding programs and other technologies for genetic improvement using Nile tilapia (Bolivar, 1993; Acosta *et al*., 2006), leading to significant advances in the genetic improvement of tilapia and development of different strains which had been sustained during the past three decades (**Box 1**). At the beginning, the main focus of most of these breeding programs was to improve the cultured tilapia's overall farm performance such as in the Fish Genetics Project of the Freshwater Aquaculture Center (FAC) of Central Luzon State University (CLSU), which produced the FAC-selected Tilapia (FaST) strain in 1986-1988, and its Genetic Improvement of Farmed Tilapia (GIFT) Project, which developed the Genetically Improved Farmed Tilapia (GIFT) strain in 1988-1997 (Eknath *et al*., 1993; Bolivar and Newkirk, 2000).

Both projects successfully produced tilapia strains which have higher growth and survival performance compared to the farmed local strain. Simultaneous with the GIFT program, YY-male and Genetically Male Tilapia (GMT) was developed using YY-male technology that was conceptualized as a form of breeding program that generates monosex tilapia (with YY genotypes instead of XY for normal males) providing an alternative to hormonal sex reversal and hybridization. After the development of GIFT, successive projects which intended to perform further enhancement of this strain were conducted and subsequently developed Genomar Supreme Tilapia or GST (Gjoen, 2001) and Genetically Enhanced Tilapia - Excellent (GET-EXCEL) strain (Tayamen, 2005).

Special breeds of tilapia that can perform well in different culture environments were also produced such as the COLD strain that can be farmed in low-temperature environments and saline-tolerant strains like BEST and Molobicus (Villegas, 1990; Romana-Eguia and Eguia, 1999; Tayamen *et al*., 2002; Rosario *et al*., 2004). At present, tilapia is the second most important food fish for domestic consumption in the Philippines, next to milkfish (Lopez *et al*., 2005; BFAR, 2006). This increase in the national demand for tilapia is a result of increased production brought about by the various efforts in tilapia genetics R&D. Over

Box 1. The "fruits" of genetic research on tilapia in the Philippines (Modified from Abella, 2006; Acosta, 2009)							
Strain Developed (Popular Name)	**Research**	**Project Year**	**Implementing institutions**	**Donor(s)**	**Significant Results**	**Producers**	**Date of Commercial Distribution**
FaST (FAC-selected Tilapia also called "IDRC" strain in local market)	Fish Genetics Project of FAC	1986-1996	FAC-CLSU	International Development Research Centre (IDRC)	Produced fast-growing strains of *O. niloticus*	Hatcheries which purchase broodstock from FAC	1993
GIFT (Genetically Improved Farmed Tilapia)	Genetic Improvement of Farmed Tilapia	1988-1997	Institute of Aquaculture Research (AKVAFORSK) of Norway, FAC-CLSU, ICLARM, BFAR-NFFTC, UPMSI	Asian Development Bank and United Nations Development Programme	Produced fast-growing strains of *O. niloticus* and demonstrated that *O. niloticus* did respond positively to selection	GIFT - Genetically Improved Farmed Tilapia)	1997
GST (GenoMar Supreme Tilapia)		1999-2002		GenoMar	Application of DNA genotyping technology, selection of differential increases, and total genetic gain for growth rate are expected to result in 40% higher performance than the ninth-generation fish	GenoMar Philippines, Inc.	2002
YY male/ GMT (Genetically Male Tilapia or sometimes called "YY")	Genetic Manipulation for the Improvement of Tilapias	1988-1997	University of Wales, Swansea/FAC-CLSU, BFAR-National Freshwater Fisheries Technology Center (NFFTC)	Overseas Development Administration (ODA)	Produced genetically male tilapia for grow out and YY breeders for fingerling production	produced by Fishgen Ltd. and by Phil-Fishgen and its accredited hatcheries in Philippines	?
GET EXCEL (Genetically Enhanced Tilapia - EXcellent strain that has a Comparative advantage over other tilapia strains for Entrepreneurial Livelihoods)		2002	BFAR-NFFTC	DA-BAR	Combining strain crosses and adopting within family selection of four different strains of *O. niloticus*	produced by NFFTC and its accredited multipliers	
BEST (or Brackishwater Enhanced Selected Tilapia)	Development of Saline and Cold Tolerant Tilapia	1998-present	FAC-CLSU, BFAR-NFFTC, University of the Philippines in the Visayas	DA-BAR	Formed a base population from four different *Oreochromis* species by combining best performing purebreds and crossbreeds after rigid evaluation in different environments	produced by NFFTC and its accredited multipliers	
Cold-tolerant tilapia Molobicus	Development of Saline Tolerant Tilapia Hybrid (Molobicus Program)	1998-present	BFAR-National Integrated Fisheries Technology Development Center (NIFTDC)	PCAMRD and Centre de Cooperation Internationale en Recherche Agronomique pour le Development (CIRAD)	Developed saline tilapia hybrids through hybridization using *O.niloticus* and *O. mossambicus*	By NIFTDC and its accredited multipliers	
SST (SEAFDEC-Selected Strain)		1999-?	Aquaculture Department (AQD) of the Southeast Asian Fisheries Development Center		Produced a fast growing strain of *O. niloticus* from modified mass selection technique with collimation technique and development of a small-farm, low-cost selection program	SEAFDEC/AQD	

the last 25 years, the tilapia industry in the country has achieved tremendous progress due to the development and production of improved tilapia strains (Tayamen *et al*., 2006).

Status and trend of tilapia production in the Philippines

With the decline in the consumption of milkfish and roundscad, tilapia has become one of the cheapest sources of animal protein in the diet of Filipinos (Edwards, 2006; ADB, 2005). Currently, the tilapia industry in the country accounts for 12% of the total aquaculture GDP. Tilapia is also the main freshwater fish species cultured in the Philippines, comprising about 79% of the total freshwater aquaculture production in 2010 (BFAR, 2010). Improved strains of tilapia that farmers can choose from include: GIFT, FaST, GST, SST, BEST, COLD, EXCEL, and Molobicus (Toledo, *et al*., 2009). After the introduction of enhanced tilapia strains, the average per capita consumption of tilapia in the Philippines increased by 474 percent, from 0.66 kg/person/year (1979-1988) to 3.13 kg/person/year in 2010 (Yosef, 2009; BAS, 2010).

The tilapia industry in the Philippines increased eminently achieving a remarkable growth of 50% from 2002 to 2012 (**Fig. 3**). More than 90% of the total tilapia production in the Philippines comes from freshwater environments, of which 40% is produced from freshwater fishponds. Supply coming from brackishwater environments has yet to generate much impact on the total production despite having salt-tolerant strains available for farming. About 80% of the country's tilapia supply comes from central Luzon area (**Fig. 4**), of which production from Pampanga contributed the highest at 39% of the total annual supply followed by Batangas (24%), Rizal (5%), Laguna (4%), and Pangasinan (3%). The improved performance of the country's tilapia industry could be attributed to the accessibility of wide range of tilapia strains and increased resources and labor force as farming operations of tilapia became widespread.

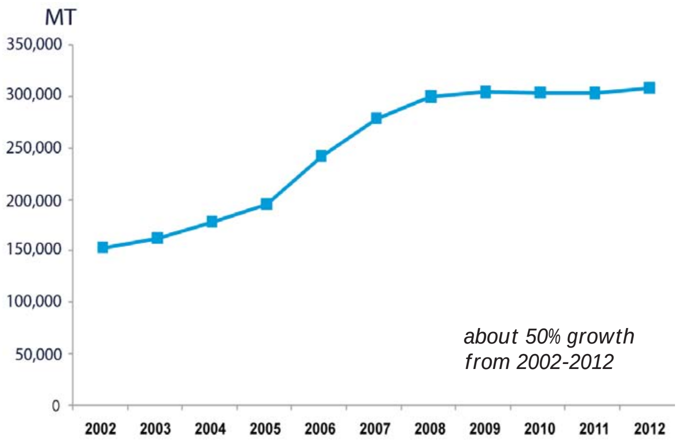

Fig. 3. Total annual tilapia production of the Philippines from 2002 to 2012 (BAS, 2012)

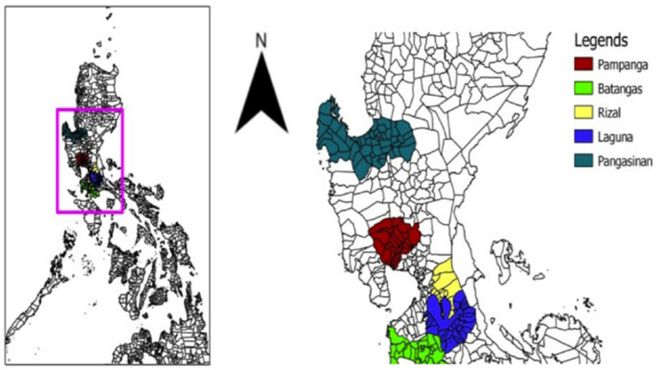

Fig. 4. Five top tilapia producing provinces of the Philippines

Moreover, in view of the increased accessibility to and availability of input supply, sustained advisory services, expanding consumer markets, rapid development of marketing channels in response to the market-driven demand, and increased availability of high performance tilapia seeds, production of farmed tilapia had tremendously increased from 1981 to 2001 (ADB, 2005). The development of genetically-enhanced tilapia from various breeding programs in the country increased the yields and kept tilapia affordable for the poor. GIFT and GIFT-derived tilapia strains comprised 68% of the total tilapia seeds produced in the country in 2003 (ADB, 2005) which validates the significant contribution of this genetic improvement to the increasing production of tilapia in the country.

In 2004, Philippines with a total production of 145,869 MT, ranked third among the top tilapia producers in the world. The ADB ascertained that GIFT and GIFT-derived strains are responsible for most of increasing tilapia production in the last two decades (Acosta and Gupta, 2009). It is important to note that tilapia production from marine or brackishwater culture areas, especially in Visayas and Mindanao has not yet been commercially significant to a great extent (ADB, 2007; Toledo *et al*., 2008). However, despite the increasing production of tilapia in the Philippines, such feat is still not as significant as those in other Asian countries due to certain obstacles encountered in the tilapia culture industry with the still nascent management and dissemination techniques (Yosef 2009).

Utilization of tilapia in the Philippines: Value-adding

Of the total tilapia supply, only a small portion is processed as value-added products due to household consumers' preference for live fish. In domestic markets, tilapia is usually sold as whole fish, either frozen or fresh but sometimes could also be available in dried and fillet forms that are supplied to major outlets such as supermarkets and other food chains. In 2002, a project of BFAR on "Value-added products from Tilapia" sought for an appropriate processing technology to create value-added products for tilapia, with the objective of increasing the economic returns from its production (dela Cruz, 2010). The project successfully developed four different products, namely: *longganisa*, nuggets (breaded tilapia), *tocino*, and rolls. In addition, another processed tilapia product is known as *tilanggit*, a small (juvenile stage), dried, and deboned tilapia similar to juvenile stage of rabbitfish known as *danggit* in the Philippines (Fernandez, 2008).

Biofactory Opportunities and Challenges for Tilapia and Milkfish

Biofactories utilized nowadays are microbial cells, plant cells, algal cells, and mammalian cells, most of which are already established biofactories that cover wide range of applications in various industries especially in agriculture and biomedical fields (Sarmidi and El Enshasy, 2012). Most biofactories are sourced primarily for bioactive metabolites including enzymes (*e.g.* amylases, glucose oxidase, cholesterol oxidase), antibiotics (*e.g.* penicillins, erythromycin, rifamycins), recombinant proteins (*e.g.* insulin, human growth hormones), and other biopharmaceuticals while other biofactories produce bioplastics and biodiesel (Sarmidi and El Enshasy, 2012). Milkfish and tilapia conform to the general advantages of fish as potential biofactories. Both are relatively cheap, easy to manage and culture, can be produced in high volume, and are renewable resources. Commercially-valued compounds known to have been extracted from fish include collagen, fish oil-derived oils, and fish protein hydrolysates.

Interestingly, these products can be extracted from fish by-products including head, skin, fins, trimmings, fins, frames, viscera and roes (Chalamaiah *et al.*, 2012). The fish processing industry has been reported to generate 60% of fish wastes and only 40% fish products for human consumption (Dekkers *et al.*, 2011). These by-products contain good amount of protein rich material that are normally processed into low market-value products, such as animal feed, fish meal and fertilizer (Hsu, 2010). In the Philippines, by-products generated from tilapia and milkfish industry, and fisheries in general, are considered as wastes and often thrown away after fish processing such as deboning and filleting. Although efforts on value-adding are also employed, these are not very extensive. With increasing tilapia and milkfish production every year, fish by-products discarded as wastes will also continue to rise. Therefore, establishing milkfish and tilapia as biofactories may become the practical alternative for fish-processing waste management while generating additional profits at the same time.

Tilapia and milkfish as biofactories for collagen

In its purified form, collagen has been used in various pharmaceutical and biomedical applications such as treatment for hypertension, urinary incontinence and pains associated with osteoarthritis; in tissue engineering for implants in humans; and inhibition of angiogenic diseases, such as diabetes complications, obesity, and arthritis (Ogawa *et al.*, 2004). In the cosmetics industry, collagen has been utilized in skin care products as humectant or moisturizing agent (Peng *et al.*, 2004). At present, collagen extracted from aquatic organisms is more preferred for human consumption than mammalian-derived collagen because currently, the main sources of collagen in many fields are limited to those of bovine or porcine dermis which pose health risks due to the outbreak of transmissible spongiform encephalopathy (TSE) and bovine spongiform encephalopathy (BSE), as well as foot-and-mouth disease (FMD) crisis (Zhang *et al.*, 2011). Tilapia has been reported to be an excellent source of Type I collagen (Ikoma *et al.*, 2003; Sujithra *et al.*, 2013), which could be collected from the skin, scales, fins, and bones of tilapia (Ogawa *et al.*, 2004; Pang *et al.*, 2013). Nevertheless, the feasibility of different milkfish parts as potential source of collagen has yet to be explored. Thus, characterization and screening should be initiated to determine its utilization in collagen production. The emerging demand for fish-derived collagen is a very potent driver to develop and establish fish biofactories for this product in the Philippines. In Southeast Asia, the University of Putra Malaysia and Bionic Lifesciences Sdn. Bhd. have already ventured into this market, establishing the first halal collagen extractor factory from tilapia fish skins and started producing aquatic collagen in commercial scale (UPM News Portal, 2011).

Tilapia and milkfish as biofactories for fish oil-derived fatty acid and biodiesel

Fish wastes, especially the viscera, are essential raw materials to produce fish oil. Representing up to 15% of the total fish body weight, fish viscera usually have no commercial value (Oliveira *et al*., 2013) but these parts are primary source of fish oil which is subsequently used to extract omega-3 and biodiesel, the other valuable biochemical products that can be potentially sourced from tilapia and milkfish. Omega-3 oil can be utilized as food supplement and to fortify various food products such as orange juice, bread, yogurt, and butter (Fitzsimmons, 2008). Fish wastes could also be used to produce biodiesel after the fish oil has been extracted and processed. Compared to petroleum diesel, biofuel from vegetable oils and animal fats is biodegradable, has non-toxic profile and creates low greenhouse gas emissions (Oliveira *et al*., 2013). Two successful companies are making significant contributions to local energy production using fish residues: Aquafina in Honduras using tilapia wastes and Agifish in Vietnam which uses catfish wastes (Piccolo, 2008). It has been estimated that Aquafina has been producing over 15,000 liters/day of biodiesel from tilapia fish oil (Piccolo, 2008). Recently, Brazil's National Department of Works Against Drought (DNOCS) has announced its planned establishment of fish waste biodiesel plants to cut down 50% of tilapia wastes while producing more than 8,000 liters of biodiesel per day (Lane, 2013). Converting tons of fish wastes from tilapia, milkfish, and other fish species into omega-3 or biodiesel is another opportunity to boost revenues of these two fish industries. One of the good news about the technology used in the production of biofuels from fish wastes is that it is transferable (Lane, 2008) adding to another reason for developing tilapia and milkfish as biofactories.

Tilapia and milkfish as biofactories for fish protein hydrolysates

Fish protein hydrolysates (FPHs) are smaller peptide fragments of usually 2-20 amino acid in length produced from the enzymatic breakdown of fish proteins (Chalamaiah *et al*., 2012). FPHs have been utilized as nutritional supplement, functional ingredients in different foods, and aquaculture feeds for enhancing the growth and survival of fish (Chalamaiah *et al*., 2012). FPHs can also be extracted from fish by-products, hence, another alternative for the utilization of increasing fish processing wastes. Protein hydrolysates from fish are currently considered as the most important source of protein and bio-active peptides which is why fish FPHs have gained great attention to food scientists and have been utilized in various industrial applications (Chamalaiah *et al*., 2012). Tilapia has been reported to be good source of desirable quality of FPH (Foh *et al*., 2011) and studies have exemplified the potential of tilapia FPHs as antioxidant agents (Raghavan *et al*., 2009; Shamloo *et al*., 2012). On the other hand, protein hydrolysates from milkfish have yet to be sufficiently characterized and documented. This is a good avenue for exploring the potential of milkfish or its by-products as a new source of high-grade FPHs.

Tilapia and milkfish as biofactories for recombinant proteins

Production of recombinant proteins requires transgenic technology to genetically alter organisms to express the desired protein. Producing recombinant proteins using transgenic animals offers a renewable source of bioactive products that are difficult to obtain by other means (Houdebine, 2000; Lubo, 2000). This "biopharming" concept (also known as "molecular farming" in plant biotechnology) is the combination of current agricultural practices and biotechnological approaches for the low-cost production of molecules of commercial value (Twine, 2005) and is considered the next major development in both farming and pharmaceutical production (Kaye-Blake *et al*., 2007).

The use of fish as biofactories or bioreactors is an emerging approach for the production of eukaryotic recombinant proteins (Zbikowska, 2003). However, fish have not been used as a biofactory (Rocha *et al*., 2003) even with the advances in the applications of transgenic technology such as growth enhancement, disease resistance, and cold resistance which have already been established in different species of fish. Using fish offers several advantages such as the large number of eggs produced and their development outside the female, which does not occur in mammals (Rocha *et al*., 2003). In addition, fish is also a good option for biofactory because of its short generation time, low cost of cultivation, easy maintenance, and its use for experimentation is more ethically acceptable than using mammalian or avian models, and there is no present evidence of the replication or transfer of prions in and from fish (Maclean *et al*., 2005; Hu *et al*., 2011).

To date, only few researches explored the use of fish as pharmaceutical biofactory, although several companies have already ventured in the development of fish as biofactory (Bostock, 1998). Calcitonin has been produced using a transgenic salmon through the initiative of DiverDrugs in Spain. Japan's Shina Canning Co. Ltd. has also produced collagen from transgenic fish. The most notable advancement in this field is the production of humanized insulin from islet cells (Brockmann bodies) of transgenic tilapia (Pohadjak *et al*., 2004). This transgenic tilapia could become a suitable, inexpensive source of islet tissue that can be easily mass-produced for clinical islet xenotransplantation to treat insulin deficiency (Pohadjak *et al*., 2004). Fish eggs from transgenic fishes have also been utilized in the production of heterologous recombinant proteins. For example, human coagulating factor VII (hCFVII), a blood clotting factor released during internal tissue injury, has been reported to be expressed ubiquitously in tilapia embryos (Hwang *et al*., 2004) while successful production of functional recombinant goldfish luteinizing hormone (gfLH) was done using transgenic rainbow trout embryos (Morita *et al*., 2004). The latter experiment which highlighted on the use of fish eggs as bioreactors has advantages including high expression of target protein at low cost and the capability of performing complex post-transcriptional modifications. Recently, Hu *et al*. (2011) used zebra-fish eggs as bioreactors to produce mature tilapia insulin-like growth factor (IGF) proteins using the oocyte-specific zona pellucida (zp3) promoter. From the 650 fish eggs, about 0.58 and 0.49 mg of purified recombinant tilapia IGF-1 and IGF-2, respectively, were extracted from the cytoplasm of the eggs. Insulin-like growth factors, especially IGF-1, promote growth by stimulating somatic growth and cell proliferation in vertebrates (Castillo *et al*., 2004). The biologically-related roles of tilapia IGFs have attracted attention among researchers in aquaculture, biomaterials and cosmetic biotechnology (Hu *et al*., 2011). In fact, there has been an initiative to incorporate IGFs as feed additive to enhance growth (Liao *et al*., 2008). These studies demonstrate that transgenic fish as biofactory or bioreactor has a great potential in the practical and commercial production of valuable therapeutic proteins.

Research in fish biotechnology has not been proliferative in the Philippines. The earliest effort in the application of fish transgenesis in the production of pharmaceutical biomolecules was made through a collaboration between the formerly-known Department of Science and Technology-Philippine Council for Aquatic and Marine Research and Development (DOST-PCAMRD) now Philippine Council for Agriculture and Aquatic Resources Research and Development or DOST-PCAARRD) and a team of Canadian scientists. Their study aimed to produce transgenic tilapia that could produce human insulin (AquaNews, 1998). Unfortunately, no subsequent studies followed through as a continuation for this project. It is also important to note that there have been no transgenic-technology applications in aquaculture after the completion of this project, which could be due to the need to address more pressing concerns such as performance improvement in the fish aquaculture sector, particularly in tilapia industry.

The research efforts had since then been focused on addressing the deteriorating quality of tilapia being farmed using classical genetic techniques while biotechnology-based experiments, such as transgenesis, were not as relevant as conducting genetic improvement programs at that period and, therefore, were least prioritized. Tilapia is one of the fish species that has attained tremendous success in terms of advanced genetic applications. Robust genetic information on tilapia can now be accessed including its whole genome sequence. Unlike tilapia, milkfish has not been subjected into intensive genetic experimentations, adding up to the piling challenges if transgenic technology is sought to be applied. In addition, obstacles that will transpire when considering research programs using transgenic fish models in general, aside from financial constraints, also include lack of facilities and limited personnel with technical expertise in the field. However, these limitations should be perceived as another opportunity to promote R&D in this field.

Conclusion and Way Forward

Reports have shown that in 2012, production of milkfish and tilapia in the Philippines accounted for 15% and 12% of the total aquaculture production of the country, respectively (Bureau of Agricultural Statistics, 2012). Production of these commodities was reported to have increased in terms of volume and value during the succeeding years. Increase in milkfish production has been attributed to the result of good farm management, availability of quality fry/fingerlings, and proper feeding practices, while increase in the country's production of tilapia has been possible due to the easy access to wide range of tilapia strains, and increased resources and labor force in tilapia farming operations. These commodities are generally consumed in the country as fresh or frozen or processed products or in modified form as value-added products. It is a fact that the sustainability of tilapia and milkfish production has been one of the paramount concerns of the country's aquaculture sector, however, profitability and competitive advantage of these industries will also have to eventually rely on new approaches that involve value-adding strategies and genetic technologies. With the availability of advanced technologies at present and a potential market for the production of bioactive compounds and molecules of commercial value, tilapia and milkfish industries could start to shift gear towards the utilization of both fish species as new and renewable source of valuable compounds, leading to the establishment of biofactories. Tilapia and milkfish are relatively advantageous because they are relatively cheap to cultivate and manage and can be easily produced in large quantities.

Associated with the increase in tilapia and milkfish production is also the increase in fish processing by-products that are usually underutilized or considered as wastes. Essentially, these biofactories will strongly depend on fish processing by-products as the main source of raw materials in the production of bioactive compounds and molecules such as collagen, biodiesel, and fish protein hydrolysates. This effort does not only entail adding revenue and value to both fish industries but also maximizing the utility of the growing volume of fish processing by-products while reducing the possible unwanted environmental impacts of these wastes. Tilapia and milkfish as biofactories for the production of pharmaceutical recombinant proteins is also a very attractive option. However, intensification of R&D in fish biotechnology is an initial but imperative requirement to allow progress in this field. In general, future research on tilapia and milkfish biofactories should therefore be directed towards the development of designs, from the cultivation of the organism to extraction and purification of bioactive compounds. In addition, if industrial platform will be established in the future, bioprocess development and complete bioprocess design are required and should be carefully considered (Sarmidi and El Ensashy, 2012).

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About the Authors

Mudjekeewis Santos, **Irma Destura** and **June Ordoñez** are researchers from the National Fisheries Research and Development Institute of the Philippine Bureau of Fisheries and Aquatic Resources with addresses at 101 Mother Ignacia St., Quezon City 1103, Philippines. Email: *mudjiesantos@gmail.com*