

## Review Article

### Global diversification of tilapia production techniques: recent

#### overview- Part 1

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#### ABSTRACT

Tilapia secure a bright position in finfish production worldwide because of their excellent flesh quality, disease resistance as well as the capacity to survive in a variety of farming systems and environmental circumstances. They hold a promising position in the propagation of aquaculture in developing countries because the culture is easy, farmers are friendly, and minimum infrastructure is required for their culture. Although more than 70 Although there are several tilapia species, only three are commonly cultivated: *Oreochromis niloticus*, *Oryza mossambicus*, and *Oryza aureus*, where Nile tilapia contributing about 75% of the total production of tilapia. accounts for approximately 8.3% of the total aquaculture production. These three species grow rapidly under diverse environmental conditions and are able to adapt to different culture systems. These qualities have made them a natural choice for freshwater aquaculture to address food security and the requirement of dietary protein in several developing countries in Asia. However, prolific breeding results in productivity loss, the non-availability of quality seeds, and improper management strategies, which have hindered their cultivation and adoption worldwide for many decades. These problems have been resolved to a large extent in recent years through the introduction of innovative and improved culture practices, better farm management strategies, and the introduction of hormonal and selective breeding techniques. This review explores how tilapia production has increased through the diversification of cultural focused techniques under diverse climatic conditions, infrastructure facilities, and the application of modern tools. Advanced genetic tools and polyculture opportunities could serve as the main target areas to provide maximum efforts for sustainable outcomes.

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What is or are the conclusions and recommendations?

*Keywords: Monoculture, Polyculture, Hormones, Genetics, Production*

#### 1. INTRODUCTION

One of the areas of food production that is expanding the quickest in the globe is aquaculture. During the last decade, the aquaculture of farmed animals industry has become a source of income for millions of people across the globe, which in turn has played a significant role in providing food security to people and in fighting against malnutrition and poverty (Bene et al. 2015, 2016; Fiedler et al. 2016; Silva et al. 2021;

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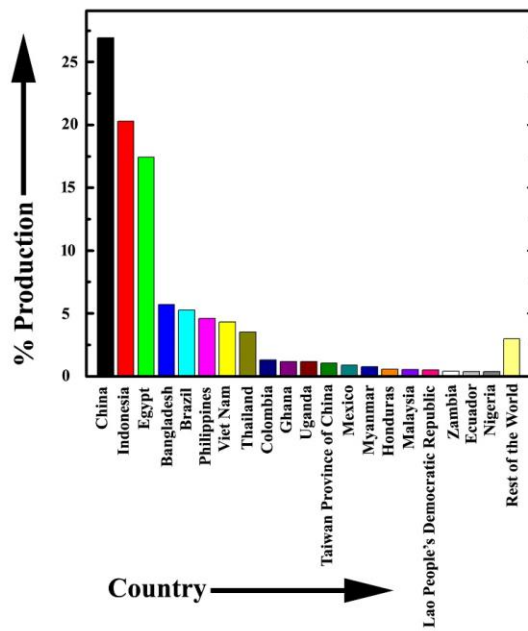
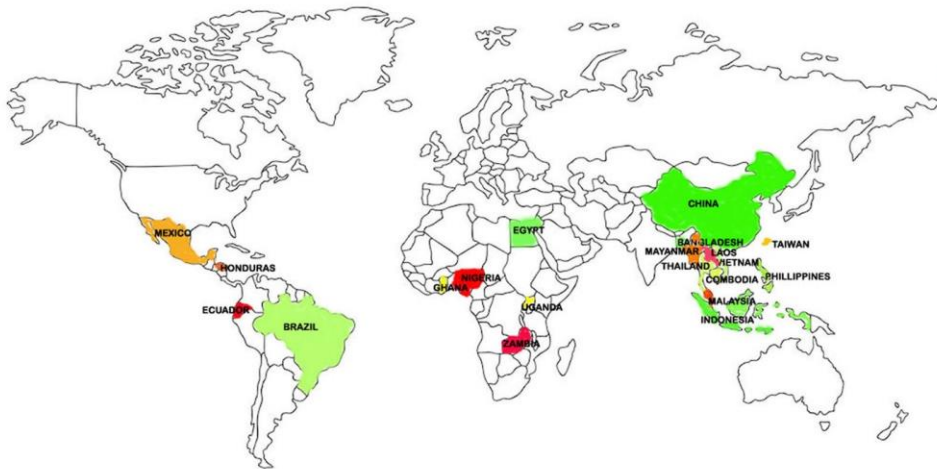
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Naylor et al. 2021). While global capture fisheries production has reached a plateau during the last fifteen years with production fluctuating between 90.4 to 96.4 million metric tons, aquaculture production of farmed animals has dramatically increased from 14.9 million metric tons in 1986-1995 to 82.1 million metric tons in 2018, inland aquaculture contributing more than 62% of the production (FAO, 2020<sup>a</sup>). Inland aquaculture production is dominated by freshwater finfish culture (47 million metric tons) compared with only 7.3 million metric tons of production from marine and coastal finfish aquaculture. The per capita intake of fish increased from 9.0 kg (live weight equivalent) in 1961 to 20.3 kg in 2017 (FAO, 2020<sup>a</sup>), and even this rapid rise in finfish production is not enough to keep up with the demand, requiring further technological advancements and cultural variety.

The inland aquaculture production of finfish is restricted to a limited number of finfish species. Tilapias secured a bright position in finfish production, with an annual production value of approximately 6.03 million tonnes in 2018. The top 20 countries based on the percentage of tilapia production are depicted in **Figure 1**.

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**Figure 1:** Top 20 most tilapia producing countries accounted for 97% of global production (FAO, 2020).

Although more than 70 species of tilapia has so far been described, the culture is restricted mainly to three species - the *Oreochromis niloticus* (Nile tilapia), *Mozambique mossambicus*, and Blue tilapia (*Oreochromis aureus* (Blue tilapia)). Nile tilapia *O. niloticus* contributes approximately 75% of the total production of tilapia (4525.4 thousand tons), sharing approximately 8.3% of the total aquaculture production in 2018 and securing third place only after grass carp *Ctenopharyngodon idella* (10.5%) and silver carp *Hypophthalmichthys molitrix* (8.8%) (Yanez et al. 2020; FAO, 2020; Miao and Wang 2020). However, global contribution of *O. niloticus* decreased from 83.4% in 1998 to 75% in 2018 due to increasing cultivation of other species of tilapia (Miao and Wang 2020) and limited capacity of growth, survival and reproduction of *O. niloticus* in saline water restricting its culture mainly in freshwater condition (Ninh et al. 2014; Yanez et al. 2020). Currently, there is a record of the culture of 23 species of tilapia including ten farm raised varieties (Miao and Wang, 2020). A significant increase in the global production of *O. mossambicus* and *O. aureus* has also been observed from 40,652 tons and 844 tons in 1998 to 53,754 tons and 3,182 tons in 2018, respectively. *O. aureus* showed feasibility for cultivation in colder regions with water temperature ranging between 8°C to 9°C, and suitable for countries with rapid seasonal and climatic changes during cultivation. Owing to its delayed sexual maturity, cultivation of this species in seasonal water bodies is becoming an additional advantage. Because of their slow growth rate, early maturation, and prolific breeding, pure strains of *O. mossambicus* have little potential

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for aquaculture. These traits also create inbreeding depression, which limits the growth of other fish species. However, due to its capacity to tolerate high salinity rural farmers of some developing countries are still continuing its cultivation in extensive and low cost culture system under brackish water condition (Ninh et al. 2014).

**Finfish aquaculture** plays a leading role in addressing malnutrition, food security, and income generation in developing countries (Wally, 2016), with uneven distribution of aquaculture development across regions and countries. Many developing countries need strong aquaculture development to feed their fast-growing population, but fail to implement it due to a lack of policies, strategies, and investment from private and public entrepreneurs. While the development of infrastructure for the culture of the majority of finfish species, including carps, requires specific habitat conditions and huge investments, tilapia can be cultured in small water bodies, even at the household level, with minimum inputs and investment. Fish grow rapidly under diverse environmental conditions and are able to adapt to different culture systems. These qualities have made tilapia a natural choice of species for freshwater aquaculture to address food security and dietary protein requirements in several developing countries in Asia, Africa, and Latin America (FAO, 2017, 2019; Prabu et al. 2019; Joffre et al. 2019; Miao and Wang, 2020). Tilapia is considered an affordable animal protein for human consumption in developing countries, and has successfully spread over 125 tropical and subtropical countries (El-Sayed, 2015; FAO, 2019) and has provided a significant livelihood option to poor and marginal farmers in these countries. Emphasis is now being placed on the diversification of tilapia culture and increase in its

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production. According to FAO's prediction, the global production of tilapia is going to increase twice within 2030 compared to 2010, thereby making it one of the front liner finfish for future aquaculture development (FAO, 2018). This review explores how tilapia production has increased through various scientifically upgraded techniques.

## 2. Culture techniques to control prolific breeding

Prolific breeding of tilapia is principally controlled through two methods: (i) monosex culture and (ii) polyculture with other species.

### 2.1. Strategies for tilapia monoculture

The uncontrolled reproduction of tilapia in grow-out ponds is a major drawback for the sustainable development of tilapia culture. Several technological attempts have been made to control such unwanted situations, the most important of which is the culture of only one sex (monosex), preferably male tilapia. It has been found that male *O. niloticus* is more profitable as it grows almost twice as fast as females because of its superior physiological capabilities, aggressive feeding behavior, and maximum investment of energy in somatic growth rather than in reproduction (Prabu et al. 2019). To make a male-only culture of tilapia, the following principal techniques were used:

1. Manual segregation of sexes
2. Environmental manipulation
3. Hormonal treatments
4. Hybridization

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## 5. Genome editing

Each technique has advantages and disadvantages (Fuentes-Silva et al. 2013; Chen et al. 2018; Felix et al. 2019). This is discussed in detail below.

### 2.1.1. Manual segregation of sexes

The successful implementation of this technique depends mainly on identifying the sex of tilapia at an early stage. Sexual dimorphism exists in the urinogenital papillae of tilapia. Female individuals possess larger papillae with a wider opening than males, which helps them to release eggs during reproduction. Only skilled farmers can identify such morphological differences between male and female tilapia even at 15 g weight (Felix et al. 2019). Segregation of sexes through manual sorting is tedious and imprecise.

### 2.1.2. Environmental manipulation

Tilapia is known for its thermostatic behavior and reproductive responsiveness to changing environmental temperatures in the post-fertilization stages (Fuentes-Silva et al. 2013, Prabu et al. 2019). This phenomenon has also been observed in over 60 different commercially important species of fish, including tilapia (Budd et al. 2015). The sex-determining mechanism in tilapia is complex and varies among species. However, numerous pure and hybrid tilapia species have shown effects of temperature on sex differentiation (Felix et al. 2019; Nduku et al. 2022). The shift of sex ratio towards either males or females has been found to depend not only on specific temperature but also on genotype, which, together with environmental factors, control sex determination (Fuentes-Silva et al. 2013; Budd et al. 2015). While Nile tilapia *O.*

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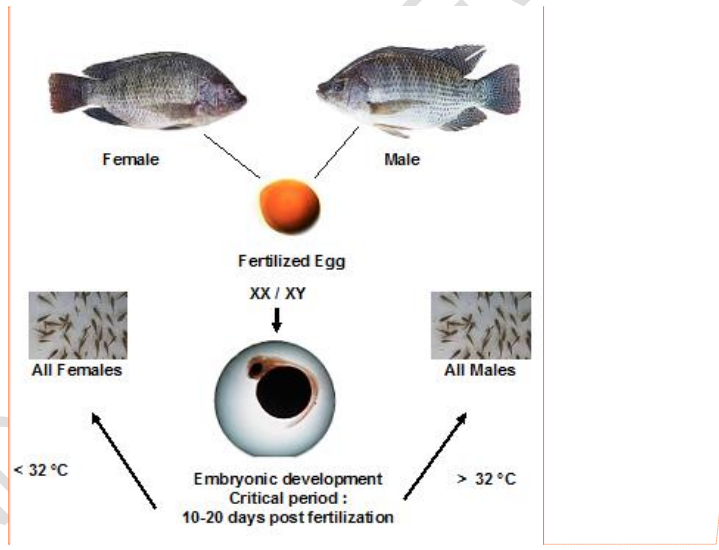
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*niloticus* is characterized by male heterogametic sex chromosomes (XY/XX), several other species of tilapia like *O. aureus*, *O. hornorum*, *O. karongae* and *Tilapia mariae* exhibit female heterogamety (ZW/ZZ) (Baroiller et al. 2009). In Nile tilapia, thermal sex determination (TSD) and environmental sex determination (ESD), more precisely, coexist alongside genetic sex determination (GSD) with male heterogamety (XY/XX) (Teng et al. 2020). Between 10 and 20 days after fertilization (dpf), there is a crucial window of time during which exposure to temperatures above 32 °C can have masculinizing effects on the developing embryo (Figure 2).

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**Figure 2.** Temperature dependent sex determining mechanism in tilapia

Angienda et al. (2010) observed that exposure to 36±0.5°C temperature for 10 or more days between post fertilization days 9–13 in *O. niloticus* embryo resulted in 86.31 % of fry as male as well as 65.25% of survival of the fry. Higher temperatures produced a higher percentage of male fry, but survival was drastically reduced. Nile tilapia is

frequently exposed to masculinizing temperatures during the critical thermosensitive period (10-20 dpf). From 0 to 9 dpf, the hatchlings spent time in the mouth of their mother, after which they were exposed to shallow marginal water with higher masculinizing temperature (32-34 °C) (Nivelle et al. 2019). However, in natural bodies, the temperature regime fluctuates between microhabitats and between day and night, which has a significant effect on the sex ratio, as is evident in blue tilapia *O. aureus*. Even in laboratory experiments, not all individuals of Nile tilapia show equal sensitivity to temperature. While some exhibit high sensitivity to temperature, giving a high proportion of males, others are insensitive to temperature, giving a balanced sex ratio, indicating an important parental effect. This is probably one of the reasons behind the limited success of temperature-induced monosex tilapia production in practical fields, despite the use of appropriate temperatures and post-fertilization periods of exposure (Bardhan et al. 2021).

Several genes influence gonadal maturation in tilapia. The *Foxl2* gene is expressed at 9dpf in XX gonads only slightly higher than in XY gonads, but the level of expression increases in XX gonads from 9 dpf onwards, but not in XY gonads. Steroidogenesis and gonadal differentiation in fish are regulated by aromatase, which is encoded by *cyp19a1a* for the gonad form and *cyp19a1b* for the brain form (Gennotte et al. 2014). *Cyp19a1a* is increased in developing ovaries of tilapia and regulates the aromatase enzyme to catalyze the conversion of androgens into 17 $\beta$  estradiol. Females become male when the *cyp19a1a* gene is inhibited, which stops the production of estrogen. The brain form of the aromatase gene (*cyp19a1b*) is also expressed very early

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in ontogenesis, but there is no difference in expression between males and females. Another gene that plays an important role in male sex differentiation is the anti-Müllerian hormone (*amh*) gene. *Amh* is upregulated in XY gonads of tilapia during 10 to 15 dpf and plays a role in testicular differentiation. Temperature application (32–36 °C) at 10–20 dpf increases *amh* gene gonadal expression while suppressing brain aromatase activity, *foxl2* and *cyp19a1a* gene expression (Poonlaphdecha et al. 2011; Gennotte et al. 2014).

### 2.1.3. Hormonal manipulation

The application of synthetic steroid hormones is considered one of the most promising and successful techniques for producing monosex tilapia. Synthetic androgen application can transform the entire population into a male-only population and exhibit all male characteristics, even if some individuals possess the female genotype (XX). Similarly, the application of ~~estrogen~~estrogen can transform the entire population into a female-only population, although some individuals may contain the male genotype (XY). Hormones are generally applied during the developmental stage, from the time of hatching up to approximately two weeks of age. At this stage, fish remain sexually undifferentiated, and the quantitative existence of the male hormone (androgen) and female hormone (estrogen) remains equal in the fish body. Depending on the hormone used at this point, the fish will be directed toward either males or females. Hormones should be administered before the gonad is differentiated, which in turn is influenced by certain genes. In Nile tilapia, expression of *cyp19a1a* is upregulated in female gonads from 9 dpf onwards (Poonlaphdecha et

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al. 2013). Treatment with estrogen increases the expression of *cyp19a1a* but decreases the expression of *amh*. Gennotte et al. (2014) observed that sensitivity of XY progeny to estrogen varied widely among individuals and YY progeny was insensitive to the hormone indicating a probable feminizing factor linked to X- chromosome only.

There are two methods of hormonal treatment.

(i) *Immersion method*: Fertilized eggs or hatchlings were immersed in a hormonal solution.

(ii) *Oral administration*: Hormone is generally administered through food at the fry stage.

Oral administration of hormones has been reported to be the most commercially applied method for the sex reversal of *O. niloticus* (Wahby and Shalaby, 2010; Celik et al. 2011). However, the immersion method is a more economically viable option (Felix et al. 2019). Table 1 presents the different hormonal agents, doses, and application methods used for sex reversal in tilapia. While the success of sex reversal varies with hormonal agents and species, the success rate also depends on the technique of application. Fadrozole, Poly lactic-co-glycolic acid (PLGA)-loaded fadrozole, and 17  $\alpha$ -methyl testosterone produced high success rates for masculinization. The oral method was found to be the most effective mode of administration of most agents for most of the species tested, except 17 was testosterone being found equally effective in oral administration and immersion methods for Nile tilapia *O. niloticus*. Seventeen  $\alpha$ -Methyl dihydrotestosterone has been tested for this species only under the immersion method and found to be 100 % effective.

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**Table 1:** Hormonal agents and their dose used for sex reversal of tilapia

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Species	Hormonal agent	Chemical class	Dose	Duration	Male (%)	Female (%)	Application method
<i>O. niloticus</i>	Fadrozole	Nonsteroidal aromatase inhibitor	75 - 100 mg/kg	30 days	100	0	
<i>O. niloticus</i>	Poly lactic-co-glycolic acid (PLGA) loaded fadrozole	Nonsteroidal aromatase inhibitor nanoparticle	300 - 500 ppm	30 days	100	0	Orally fed
<i>O. niloticus</i>	17 $\alpha$ -methyl testosterone		70 mg/kg	25 days	98	2	
<i>O. niloticus</i>	17 $\alpha$ -methyltestosterone		1800 $\mu$ g/L	8 h	98.4	1.6	Immersion
<i>O. niloticus</i>	17 $\alpha$ methyl dihydrotestosterone	Synthetic steroid hormone	1800 $\mu$ g/L	4 hours	100	0	
<i>O. niloticus</i> $\times$ <i>O. aureus</i> hybrid	17- $\alpha$ -methyl testosterone		60 mg/kg	25-28 days	$\geq$ 96	$\leq$ 4	
<i>O. mossambicus</i>	17 $\alpha$ -methyl testosterone		75 mg/kg	For 21 days	98.09	1.91	
<i>O. niloticus</i>	17- $\alpha$ - methyl testosterone		60mg/kg	28 days	94.44	5.56	
<i>O. mossambicus</i>	Diethylstilbestrol (DES)	Nonsteroidal synthetic estrogen	>100 $\mu$ g/g diet	11-15 days	0	100	
<i>O. mossambicus</i>			50 ppm	30 days	0	100	Orally fed
<i>O. aureus</i>	Trenbolone acetate (TBA)	Synthetic growth promoter	25-100 mg/kg	28 days	98	2	
<i>O. aureus</i>	17-alpha-methyl testosterone (MT)		60 mg/kg	28 days	88.7	11.3	
<i>O. aureus</i>	Ethinyltestosterone (ET)	Synthetic steroid hormone	60-240 mg/kg	22 days	100	0	
<i>O. aureus</i>	Methyltestosterone (MT)		60-120 mg/kg	22 days	96-99	1-4	

However, the application of hormones in aquaculture production has often been debated by researchers due to their potential health hazards in humans (carcinogenic and endocrine disorders) and their residual effect on water quality, which affects aquatic biodiversity and human food security (Wang and Lu, 2016; Azizi-Lalabadi and Pirsahab, 2021). Accordingly, the use of anabolic steroids (including 17 $\alpha$ -MT) has been banned in some Asian countries, the EU, and the USA.

In India, the regulatory framework began in 2009 to monitor monosex tilapia production along with strict vigil on unofficial and unrecognized entry and trade of tilapia to ensure bio-security and quarantine management (Menaga and Fitzsimmons, 2017). To avoid risk factors for human health hazards, scientists rely on genetic modification through marker-assisted selection (MAS), crossbreeding, and sex reversal to produce all male populations worldwide (Chen et al. 2018).

#### 2.1.4. Hybridization

Hybridization is the process of cross-breeding between two species or between two strains of the same species (also known as line crossing or strain crossing) to produce sterile or all male populations (Mbiru et al. 2016; Mapenzi and Mmochi 2016; Snake et al. 2020; Goni et al. 2020; Mtaki et al. 2021). **Table 2** presents the successful interspecific hybrids of tilapia, which were predominantly male.

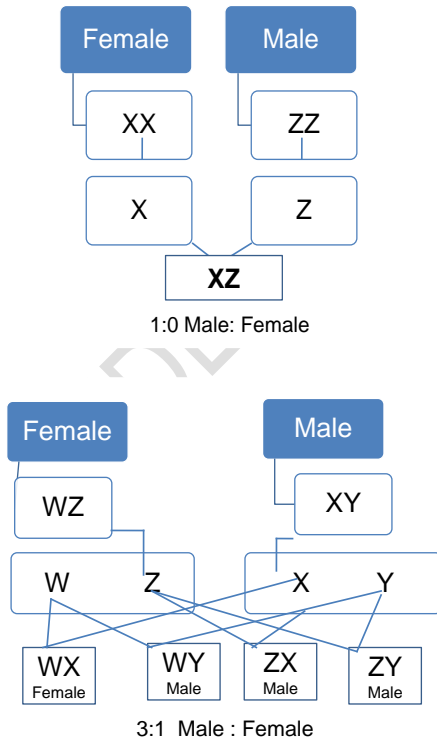
**Table 2:** Records of successful production of male tilapia hybrids

Male species (♂)	Female species (♀)	Hybrid
<i>O. aureus</i>	<i>O. niloticus</i>	<i>O. aureus</i> × <i>O. niloticus</i>
<i>O. urolepishornorum</i>	<i>O. mossambicus</i>	<i>O. urolepishornorum</i> × <i>O. mossambicus</i>
<i>O. mossambicus</i>	<i>O. aureus</i>	<i>O. mossambicus</i> × <i>O. aureus</i>
<i>O. mossambicus</i>	<i>O. spilurusniger</i>	<i>O. mossambicus</i> × <i>O. spilurusniger</i>
<i>O. aureus</i>	<i>O. niloticus</i>	<i>O. aureus</i> × <i>O. niloticus</i> (Stirling strain)
<i>O. niloticus</i>	<i>Tilapia zillii</i>	<i>O. niloticus</i> × <i>T. zillii</i>
<i>O. urolepishornorum</i>	<i>O. niloticus</i>	<i>O. urolepishornorum</i> × <i>O. niloticus</i>
<i>O. urolepis</i>	<i>O. niloticus</i>	<i>O. urolepis</i> × <i>O. niloticus</i>
<i>O. karongae</i>	<i>O. shiranus</i>	<i>O. karongae</i> × <i>O. shiranus</i>
<i>O. urolepisurolepis</i>	<i>O. niloticus</i>	<i>O. urolepisurolepis</i> × <i>O. niloticus</i>

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Inter-specific hybridization of tilapia between species with different sex-determining mechanisms, such as XY/XX and WZ/ZZ, predominantly produces male-progeny (Cnaani, 2013; Fuentes-Silva et al. 2013; El-Zaeem and Salam 2013). *O. aureus*, *O. urolepishornorum*, and *O. karongae* are members of the WZ/ZZ mechanism, whereas *O. niloticus* and *O. mossambicus* are members of the XY/XX sex-determining mechanism (Cnaani, 2013). Theoretically, 1:0 (male:female) and 3:1 (male:female) progenies are expected to be produced from crosses between female homogametic (XX) and male heterogametic (WZ) crosses and male homogametic (ZZ) and female heterogametic (WZ) crosses (**Figure 3**).

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**Figure 3:** Expected sex ratio of male (M) and female (F) offspring from hybridization between pure homogametic female and male and between heterogametic female and male

However, El-Zaeem and Salam (2013) obtained 3.59:1.00 and 4.28:1.00 (male:female) progeny after hybridization between female *O. niloticus* (XX) and male *O. aureus* (ZZ) and female *O. aureus* (WZ) and male *O. niloticus* (XY), respectively. The sex-determining mechanism of *O. shiranus* is not known. Snake et al. (2020) obtained 88 % male offspring from a cross between *O. karongae* males and *O. shiranus* females, indicating that *O. shiranus* belongs to the XX/XY mechanism because of a deviation from an expected sex ratio (1:1) in the progeny. However, hybridization did not result in 100% production of all male offspring. The lack of sex-linked markers in tilapia makes it difficult to properly understand the genetic mechanism behind sex determination, and frequent deviations from the expected sex ratio in hybridization experiments are difficult to explain using simple monofactorial sex chromosome-linked sex determination.

Of the 77 tilapia species, *O. niloticus*, *O. mossambicus*, and *O. aureus* are globally considered the most important species utilized for all male offspring production through hybridization (Felix et al. 2019). Hybrids of *O. aureus* and *O. niloticus*, apart from being predominantly male tilapia, render some qualitative improvements in terms of growth performance, fillet yield, and disease resistance (Chen et al. 2018). However, as mentioned above, the production of 100% pure male hybrids has yet to be achieved. Moreover, the maintenance of pure brood stocks, development of special hatcheries, and employment of skilled laborers make the

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process expensive (Fuentes-Silva et al. 2013; Bardhan et al. 2021). Thus, hybrid production is not a sustainable solution for male tilapia production for small and marginal farmers in rural areas in developing countries, and the global production of *O. niloticus* × *O. aureus* hybrid is considered to be the third most important variety of farmed tilapia, with a total production of 406,048 tons in 2018, accounting for 6.7% of total farmed tilapia production worldwide. Hybridization of *O. mossambicus* with *O. niloticus* produced red tilapia hybrids. Such attractive phenotypical characteristics, salinity tolerance, and high growth potential have made red tilapia a preferred cultivable fish species, especially for brackish water cultivation (Haque et al. 2016; Mtaki et al. 2021). Recent evidence indicates that a low-input farming system is sufficient for red tilapia culture, thereby making it a potential candidate for culture by marginal rural farmers (Joffre et al. 2019).

#### 2.1.5. Genome editing

Genome editing is a promising tool for monosex tilapia production. The CRISPR/Cas9 technique was first applied in *O. niloticus*, followed by other genetic engineering tools (Li et al. 2021). TALENs and CRISPR/Cas9 have been successfully applied by targeted mutagenesis to understand the genetic basis of sex determination and sex differentiation in *O. niloticus*. Homozygous mutations in the *amhy*, *amhrII*, and *gsdf* genes successfully reversed the male population into females, and this phenomenon in the *fox2* or *cyp19a1a* genes successfully reversed all females into males (Zhang et al., 2017). However, genetically manipulated stocks are often found inferior to wild stocks, resulting in susceptibility of tilapia to parasitic infection and disease and poor

growth, resulting in economic loss to farmers in different countries from the Asian, African, and American continents (Samaddar 2022)

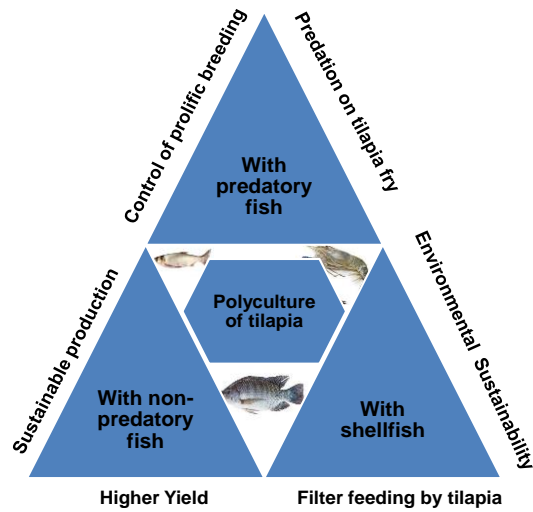
## 2.2. Strategies for tilapia polyculture

One of the most important issues related to tilapia cultivation is the huge requirement for supplementary feed, which not only makes tilapia cultivation expensive but also creates water pollution due to decomposition of the unutilized feed, particularly in monoculture systems. Such conditions create environmental stresses on fish and cause disease outbreaks. Scientists have attempted to overcome this problem through the polyculture of tilapia with other finfish and shellfish for better environmental management, profitability of the farming community, and sustainable development of aquaculture (Zhang et al. 2013; Wang and Lu, 2016; Thomas et al. 2020; Yanez et al. 2020).

As a result, the monoculture of tilapia is now largely replaced by the polyculture ~~of tilapia with other species,~~ particularly among small and marginal farmers worldwide. Since tilapia has a relatively shorter growth period (approximately 6 months to reach 500 g body weight in Nile tilapia) as compared to other finfish and shellfish species, prolific breeding and different pattern of niche utilization, success of polyculture of tilapia with other species depends upon finfish and shellfish species with which tilapia is combined, main targets of the culture as well as the farming techniques (pond based / cage / tanks / raceways etc.) adopted (Shrestha et al. 2018; Hisano et al. 2019; Arumugam et al. 2023). The main objectives of polyculture of tilapia with other species are (i) control of prolific breeding of tilapia, (ii) sustainable

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yield, and (iii) sustainable utilization of the environment. Accordingly, the three types of polyculture of tilapia with other species are as follows: (i) polyculture with predatory fish, (ii) polyculture with non-predatory fish, and (iii) polyculture with shellfish (**Figure 4**).



**Figure 4:** Objectives of polyculture of tilapia with other finfish and shellfish

### 2.2.1. Polyculture with predatory fish

The main objective of culturing tilapia with predatory fish is to control prolific breeding of tilapia. *Clarias gariepinus* and *Cichlasoma urophthalmus* are effective candidate predatory finfish species to control overcrowding of Nile tilapia (Hernandez et al. 2014; Shoko et al. 2016). Chithambaran (2019) reported that polyculture of

Sabakitilapia (*Oreochromis spilurus*) with *Lates calcariferis* ideal to control the prolific breeding and population explosion of tilapia in culture ponds.

#### 2.2.2. Polyculture with non-predatory fish

The main objective of polyculture of tilapia with other non-predatory species is to exploit the environment for sustainable yield. Efforts have been made to culture tilapia with carp species *Labeorohita*, *Cirrhinus mrigala*, *Hypophthalmichthys molitrix* and *Cyprinus carpio*. However, farmers do not prefer to include tilapia in carp culture ponds because they are poor competitors of tilapia, which causes overcrowding of the pond, resulting in poor growth of carps (Shrestha et al. 2011). Rather, cage culture of *O. niloticus* in carp-culture ponds is profitable for rural farmers (Mandal et al. 2011). The inclusion of *Tor putitora* in polyculture ponds with carp and tilapia resulted in improved yield because of selective predation of *T. putitora* on the fry of tilapia (Shrestha et al. 2011; Shrestha et al. 2018). In India and Bangladesh polyculture of tilapia with carps has been found as more profitable in comparison to monoculture in rural areas (Menaga and Fitzsimmons 2017; Ferdoushi et al. 2019). In addition to carp (*Cyprinus carpio* and *Hypophthalmichthys molitrix*), the culture of tilapia with other varieties of fish has also proven successful. The species which have been successfully used in such culture include *Mugil cephalus*, *Liza ramada*, *Chanos chanos*, *Puntius gonionotus*, *Lateolabrax japonicas*, *Colossoma brachypomus*, *Pseudosciaena crocea* and various catfish (Tahoun et al. 2013; Wang and Lu, 2016; Sudirman et al. 2020).

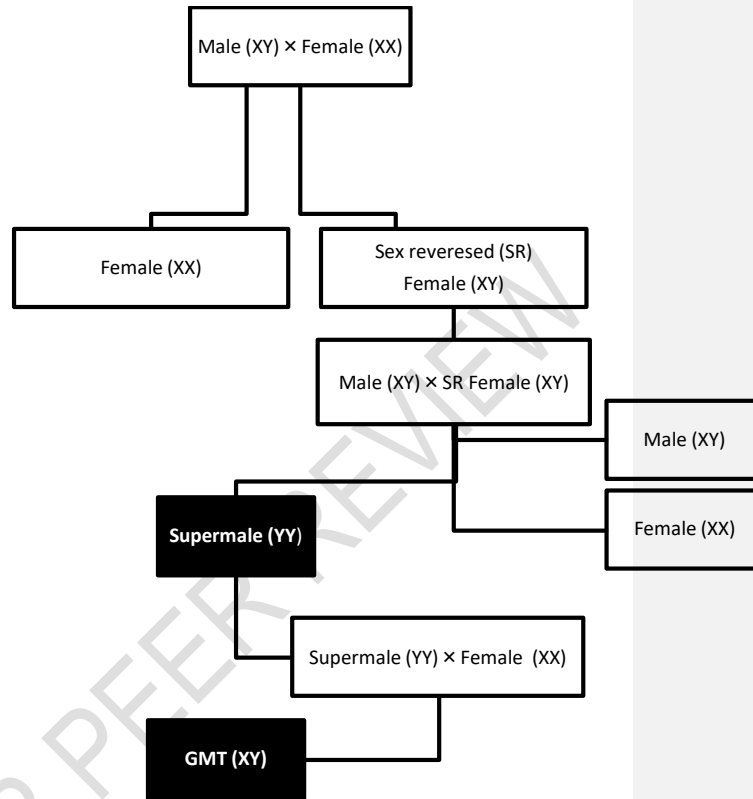
#### 2.2.3. Polyculture of tilapia with shellfish

Cultivation of tilapia along with high-value aquatic products, such as prawns and shrimp, is gaining popularity for balanced utilization of the environment. Omnivorous tilapia consume unwanted planktons and algae and reduce biological oxygen demand (BOD) in the system, while prawns and shrimps efficiently utilize benthos (Hisano et al. 2019; Arumugam et al. 2023). Farming of tilapia and shrimp together enhances shrimp health and the profit margin of farmers (Yuan et al. In 2010, Hernandez-Barraza et al. 2012). Some GIFT varieties have been found to reduce the load of luminous bacterial populations and increase shrimp survival. Intensive polyculture of *Penaeus chinensis* with Taiwanese tilapia hybrids (*O. mossambicus* × *O. niloticus*) at stocking densities of 60000 nos. and 400 kg/ha, respectively, showed improved shrimp growth performance and better utilization of the pond environment. In tropical countries cultivation of *O. niloticus* and *Macrobrachium rosenbergii* together have been recommended for better economic returns. More than 60% of shrimp farmers in the Philippines introduced tilapia into such culture systems for better economic gain. The introduction of GIFT tilapia, along with prawns, in composite carp culture systems yielded higher economic returns (Khan et al. 2016; Tran et al. 2021). In Mexico, polyculture of *O. niloticus* and crayfish (*Procambarus acanthophorus*) has shown promising results for sustainability, income generation, and environmental use in tilapia culture in rural areas (Hernandez-Vergara et al. 2018).

### 3. Genetic manipulation strategies

Commented [T39]: What word is this?

Owing to the increasing demand for tilapias on a global scale, it is necessary to obtain higher productivity in a short time span for quantitative and qualitative improvement. Genetic improvement is considered a powerful and economically viable tool for enhancing the efficiency of aquaculture industries (Yanez et al. 2020). Selective breeding is an ideal genetic stock improvement technique for tilapia (Gjedrem 2012; Murphy et al. 2020). The concept of developing “YY super male” emerged in eighties. This was reshaped by developing genetically male tilapia (GMT). A strain of Nile tilapia known as GMT is created by mating normal XX females with YY-male genotypes, resulting in all or almost all male (XY) offspring. Compared with other male fish species, GMT is said to develop faster, mature later, and produce more. Additionally, they could raise commercial yields in any setting, including ponds, where GMT has been demonstrated to produce noticeably higher yields than other strains (Wang and Shen, 2019) (Figure 5). Chen et al. (2018) applied sex linked markers during production of super males and crossing XX females with these YY males (100% progeny) for production of GMT. This technology has opened up a new horizon for all-male tilapia production on a commercial scale, but requires skill and knowledge of selective breeding.



**Figure 5:** Scheme of GMT Production

Approximately 30 years ago, Nile tilapia were produced through selective breeding and by developing Genetically Improved Farmed Tilapia (GIFT) by the WorldFish (formerly, ICLARM / International Center for Living Aquatic Resources Management,) in partnership with other global institutes (Tran et al. 2021). GIFT technology proved to be one of the most promising technologies for the commercial tilapia industry due to its positive impacts on growth, production enhancement,

reduction of culture period, and economic gain worldwide, along with harnessing local market availability and affordability (Tran et al. 2021).

The genotype  $\times$  environment interaction (G $\times$ E) plays an important role in the development of new traits and genetic improvement in tilapia, although a few authors have argued that the G $\times$ E interaction has no significant effect on body trait development in *O. niloticus* (Khaw et al. 2012; Trong et al. 2013). The best strain in one environment is likely to be the best in most or all environments (Eknath and Acosta, 1998). A high G $\times$ E interaction leads to new trait development and selection of better traits (Ansah et al. 2014). World Fish has been striving for the last two decades to develop GIFT varieties based on the environmental suitability and economic viability of different tilapia strains in different regions of the world. In India, the GIFT tilapia known as “Chitralada” was developed at Central Institute of Freshwater Aquaculture (CIFA) in Bhubaneswar in coordination with WorldFish Center breeding programme at JITRA, Malaysia (Singh and Lakra 2011). “Chitralada” is now one of the most popular GIFT Nile tilapia strains in India. However, the seed availability of this strain is limited due to restrictions imposed by the Government of India, and only a few private and state government hatcheries in the country are authorized to produce its seeds. Besides India, “Chitralada” has also been established with commercial success in Brazil (FAO, 2019). The Akosombo” strain of GIFT was successfully developed in Ghana in collaboration with the Aquaculture Research and Development Center (ARDEC) and the World Fish Center in 2003 (Ansah et al. 2014). Globally, China remains the largest producer of farmed tilapia, with a

production value of 1.62 million million tonnes in 2018 (Mioa and Wang 2020; FAO, 2020). While global position of Thailand and Philippines in GIFT production declined from top four to sixth and eighth positions contributing 3.5% and 4.6% of global production respectively, Bangladesh has developed much in GIFT production to secure fourth place with a total production of 0.34 million tonnes in 2018 (Mioa and Wang 2020; FAO, 2020).

#### **4. Recent developments in Tilapia farming processes**

The culture of tilapia in small ponds (approximately 25 to 1 ha) for a period of 5 to 10 months is simple and affordable for small and marginal farmers (FAO 2017, 2020, 2021; Nasr-Allah et al. 2021). Farmers of the developing countries of Asia, Africa, and Latin America have quickly adopted pond based culture of tilapia to meet the increasing demand for the fish. Both mono- and poly-cultures of tilapia are practiced in these countries under extensive, semi-intensive, and intensive culture systems, with an average production of 1.5 to 5 tonnes, 7.5 to 15 tonnes and 40 to 50 tons, respectively.

Cage culture is the second most popular technique for tilapia culture after pond based culture. Some of the major advantages of cage culture include better water circulation, environmental quality maintenance, better monitoring and management flexibility, low cost, high-density culture, ease of harvesting, and profitability (Prabu et al. 2019). Farmers in Cambodia, Laos, and Vietnam have commercialized cage cultures of tilapia in rivers by stocking 30-40 g size tilapia in cages and harvesting approximately 1 ton of tilapia per cage within a short time period, earning

approximately 1000 USD per cage (Bhujel, 2019). In Bangladesh, the culture of tilapia in floating cages in rivers and lakes has become popular ( Moniruzzaman et al. 2015). These small and marginal farmers are now earning about 200 USD per cage after 6 months of tilapia culture in floating cages (6 m × 3 m × 1.5 m) with a stocking density of 37-40 fish per m<sup>3</sup> (20 g average body weight) (Bhujel, 2019). Recently, periphyton-based floating cage culture of *O. niloticus* has gained popularity because of its economic viability and easy management practices (Garcia et al. 2016, 2017).

Intensive culture of tilapia in tanks and raceways is an alternative to ponds and cages in arid and semi-arid areas, where the availability of sufficient water is an issue for fish production. However, such a culture requires high investment costs, a high degree of environmental and quality control, and constant monitoring, yet offers some risk factors, such as disease outbreak and mechanical and electrical failure (Prabu et al. 2019).

Despite the huge technical improvements and success of tilapia culture in ponds, tanks, and raceways, increasing market demand necessitates better productivity, profitability, sustainability, and environmental management, which can be achieved only through the Recirculatory Aquaculture System (RAS) (Boyd et al. 2020). Worldwide, several RAS models have been standardized for the production of different tilapia varieties, depending on the environmental and socioeconomic conditions of the region. A cost-effective automated recirculation aquaculture system (ARAS) was developed by Soto-Zarazúa et al. (2011).

Recently, biofloc technology (BFT) has been used to successfully culture tilapia. It utilizes heterotrophic bacterial populations to assimilate unmanageable dissolved ammonia-nitrogen, generated in an intensive system of fish culture, and converts them into protein-rich feed for utilization by fish (Wasave et al. 2020). Tilapia is the most preferred species in the BFT-based culture system, where high stocking density of tilapia, along with maintenance of optimum water quality parameters, substantially improves production and reduces the cost of production by reducing the application of supplementary fish feed (Abduljabbar et al. 2015; Gallardo-Collí et al. 2019).

Aquaponics is an integrated food production system, in which plants and fish are grown together in the same water. The plants grow at the expense of the nitrogenous waste products released by the fish, thereby recycling the nutrients and allowing the fish to grow in a stress-free environment (Ani et al. 2022). Liang and Chien (2013) observed in a tilapia-water spinach raft aquaponics system that increased the frequency of feeding and day length could increase the production of both tilapia and the plant. An intermediate stocking density of approximately 300 fish/m<sup>3</sup> produced the best results for the growth of *Oreochromis niloticus* and lettuce (*Lactuca sativa*) in the aquaponics system (Sabwa et al. 2022).

## **5. Conclusions**

Tilapia has now become a popular finfish for culture worldwide and to address the issues of malnutrition and food security in many countries, particularly developing ones. Culturing tilapia is relatively easy, and farmers are friendly, enabling farmers of

developing nations to adopt tilapia culture widely as their livelihood. Despite the promising expansion of culture techniques and development of high-yielding varieties, some constraints still hamper the expansion of tilapia culture on a global scale. These include non-availability of quality seeds, deterioration of genetic quality, environmental degradation, and high investment required for intensive culture in tanks, raceways, recirculatory systems, and aquaponics. Such challenges enlighten further research to develop clear strategic plans for tilapia culture based on the environmental and socioeconomic conditions of a region. Advanced genetic tools and polyculture opportunities could serve as the main target areas to provide maximum effort for sustainable outcomes.

**Commented [T40]:** What is the conclusion on the techniques and strategies you discussed in the paper?

And what are recommendations

#### **CONSENT**

Not applicable

#### **ETHICAL APPROVAL**

Not applicable

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