Transgenic Fishes and their Characteristics

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Abstract

Making a genetically modified organism is called transgenesis. Fishes act as excellent models compared to mammals due to their breeding procedure, high fecundity. The main objective of gene manipulation in fishes is to enhance growth and efficiency of food conversion, tolerance to environmental variables such as temperature, salinity, to develop new color varieties. The traditional selective breeding had a huge impact on fish production but it is time consuming. Transgenic approach should supplement the selective breeding technology. Warm water fishes such as tilapia, carps and cold-water fishes such as salmon, trout were developed transgenetically. The growth in these fishes is increased by transferring growth hormone gene. Using gene manipulation, it is possible to change the phenotype and modify the cells which help in curing number of abnormalities. The modification of the fish genomes brings disease resistance, increase in growth helping in aquaculture. In the present study a transgenic fish and their characters are discussed.

Introduction

Animal that contain in it's genome, a gene that is introduced by means of transfection is called transgenic animal. The gene that is introduced is called transgene. Transfection is a process where a segment of DNA is introduced either directly or first it is integrated into a vector and then introduced into animal cell. In case of stable transfection, the introduced genes are established in the animal cells and expressed in the animal cells. The objective of producing a transgenic animal or cell is to obtain protein encoded by the transferred gene. The introduction of the gene should be aimed at improving various characters or traits such as milk, meat, wool etc., production. These organisms into which genes are transferred and made to express are called bioreactors and the process is called molecular farming. The transfer of gene also utilized in eliminating the defective genes helping in gene therapy in which number of genetic disorders is cured.

In most of the laboratories transgenic animals are routinely used as models for biomedical research. The most commonly used experimental models are genetically modified rodents. They are used in understanding gene function in relation to disease susceptibility or in determining response to therapeutic drugs. Human antibodies which are commonly used as therapeutics are produced in genetically modified mice. Large quantities of complete human proteins that are used in treatment of human diseases are produced in transgenic farm

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animals. These therapeutic proteins are synthesized in mammalian cell reactors, but these experiments are very expensive coasting over us $ 500 million. Therefore it is cheaper to produce recombinant proteins in the milk, blood and eggs of transgenic animals. The progress in these areas of research is slow.

Two bio-medical products that are synthesized in transgenic animals have received regulatory approval. Human anti-thrombin III which is therapeutic protein is the first recombinant protein that is produced in transgenic goats. This protein is used in preventing clots in patients who are having hereditary antithrombin deficiency especially when they are undergoing surgery or child birth. Another recombinant protein is human C 12 esterase inhibitor which was produced in milk of transgenic rabbits. This protein is used in treatment of hereditary angiodema which is responsible for expanding of blood vessels and cause swellings in skin.

First recombinant proteins are successfully expressed in bacteria and yeast. Afterwards large number of recombinant proteins cannot be produced in bacteria and yeast. The important reason is human recombinant proteins undergo post translational modifications in bacteria and yeast and it is completely different from the mechanism which takes place in human cells. Bacteria and yeast hosts do not act as systems which support folding of human recombinant proteins (Demain & Vaishnav, 2009). Due to this situation researchers developed alternating expression system for proper post – translational modifications of RP. These systems include transgenic animals and mammalian cell systems. The first transgenic mammal produced by microinjection of genetically engineered construct into pronucleus of mouse and zygote was formed (Hammer et. al., 1985). Later on transgenic animals are considered to be most reliable models which support production of recombinant proteins and monoclonal antibodies. Mammalian cell cultures such as Chinese Hamster Ovary (CHO) also are utilized in the production human recombinant protein.

In the past decades the global fish production is rapidly growing and main source of food yield. There is notable enlargement of aquaculture and fisheries production in 21st century (FAO, 2022). There is sharp increase from 5.2 million to 62.7 million tones in aquaculture production. Over the past 3 decades the capture fishery production increased from 68.8 million to 92.8 million tones. There is marked increase in the production of food fish and seafood which is leading to reduction in price of the fishes. Due to these high levels of investment is required in new technologies in this fishery industry (FAO, 2022; Boyd et.al., 2020). Global climate change is emerging as a challenge for sustainable aquaculture. Due to this there should be change in our management practices as the climate change is inducing changes in physical and biological conditions of fisheries. In this regard the genetically modified aquatic organisms could benefit the increase in aquaculture which can provide nutritious food for global human population and also can provide remedy for inherited diseases in aquaculture (FAO, 2022).

The nutritional security to 8 billion populations is provided by the fisheries and aquaculture sector worldwide. By 2030 to meet growing demand 70 % expansion is required in global agricultural production and associated fields. Since independence there is increase in fish production. Fish biologists are applying genomics and genetic engineering in teleost fishes.

Fast growing super fish stocks are developed successfully under laboratory conditions in the felid of aquaculture. Growth and efficiency of food conservation, tolerance to environmental factors such as temperature, salinity, resistance to pathogens, new colour variants in ornamental fishes are some of the important reasons for gene manipulation. It is comparatively less time consuming than conventional selective breeding. The first transgenic fish was produced in 1991 and as of 2003 world- wide 35 species of fishes have genetically engineered. The fishes that are generally used in genetic engineering and divided into two main groups i.e. animals that are used in culture fishery and fish that are used as model species in basic research (Chen, 1998). Carps (*Cyprinus sp.,*), Tilapia (*Oreochromis sps.*,) Salmon (*Salmo sps.*,) Cat fishes (*Ictalurus punctatus*) and Gold fish (*Carassius auratus*) are generally used in basic research. High gross food conversion, increase in weight per unit food are observed in transgenic fishes when compared to their unmodified relatives (Martinez et.al., 2000).

Microinjection, electroporation, sperm mediated gene transfer were generally used in production of transgenic animals. The fish genome is integrated with foreign gene which is inherited to their progeny in which it is expressed. Rainbow trout (*Onchorthycus myleirs*) (Maclean et. al., 1992) and gold fish (Zhu et. al., 1985) are the first reported transgenic fish in aquaculture. The growth rate is many teleost fishes is increased by targeting the gene for growth hormone. The application of transgenesis is high in ornamental fishes by targeting expression of green fluorescent protein (GFP) and Red Fluorescent Protein (RFP) (Schimizu and Schimizu 2013; Barman et. al., 2015).

The main objective of this study is to understand strategies that are used in transgenic fish production and their impact on environment and their applications in aquaculture.

Transgenic Food and Ornamental fishes – Global and Indian Scenario

Genetic engineering and gene manipulation studied in fishes is emerging as potential transforming technology. Transgenic experiments were conducted on various species of fishes. Some of the important fish groups that are utilized in gene transfer experiments are Rainbow trout, Gold fish, common carp (Aliye, 2003). Some fishes such as Japanese medaka (*Oryzias latipes*) and Zebra fish (*Brachydania rerio*). These fishes lay large number of eggs and complete their life cycle in 3 months from hatching to maturity. Their eggs are large with chorion which is semi-transparent. These characters make these fishes suitable for transgenic experiments. Number of fishes like *Misgrunus angullicauditis* (Agnes et. al., 2002), top minnows (Bartley et. al., 1986) have been genetically engineered. In the field of ornamental fishes also rainbow zebra fish have been developed transgenetically (Gang Wu et.al., 2003). Transgenic experiments have been performed on farm fishes such as gold fish (Chatkondi et. al., 1995) common carp, rainbow trout (Chourrout et.al., 1986).

Rainbow trout (*Onchorhycus mykiss*) and ornamental gold fish (*Carassius auratus*) were first transgenic food fish that were generated. Only single transgenic fish i.e. AquAdvantage Salmon is generated and approved by FDA. Zebra fish (*Danio rerio*), Medaka (*Oryzias latipes*) gold fish (*C. auratus*) are the fishes that are extensively utilized in biological studies (Clifford 2014; Gong et al., 2003; Ye et.al., 2011). Trout, carp, salmon, tilapia are few farm fishes in which gene transfer experiment are conducted keeping in view enhancement in their production which is the main objective of aquaculture. Trout and salmon earn high foreign exchange while other fish are an important source of protein. Around the world more than 50 lakhs are actively engaged with transgenic experiments on teleost fishes.

Genes and their Promoters in Transgenic Fishes

The genes such as growth hormone (GH), metallothionein (mt), antifreeze protein (AFP) are used in transgenic fish research (Gong et.al., 2003). Gene expression is stimulated by targeting the promoter sequences of heat shock protein (HSP) myosin light polypeptide chain 2 (mylz 2), keratin and metallothion (mt) successfully (Asaduzzman et. al.,2013). Tissue specific promoters like heat shock proteins 70 (HSP 70) were induced so that fishes can with stand stress conditions (Halloran et. al., 2000). Heavy metal contamination such as cadmium, mercury, zinc and copper can be detected by certain fishes because of their metallothionein promoter (Mao et. al., 2012; Ren et. al., 2006). Medaka is used to monitor estrogenic vitellogen (vtg) gene promoter for understanding reproductive events (Zeng et. al., 2005). In the field of ornamental fishes which have huge demand in the market several research organizations are developing genetic modified fishes through transgenic experiments. Ornamental fishes are developed by targeting seleted colour genes and tissue specific promoters to express the colour genes. Using tissue specific promoter gene such as krt 8 and mylz 2 that are linked to color genes such RFP, GFP, YEP and cyp were generated in Zebra fishes (Gong et. al., 2003). This has resulted in six attractive fluorescent colour varieties which are called as “GloFish” with names as cosmic blue, electric green, starfire red.

Research in India is reported since 1980. Madurai Kamaraj University (MKU) generated first transgenic fish with growth hormone (GH) gene construct. ICAR has taken number of initiatives to develop transgenic species in Indian carps such rohu, catla, zebra fish. Several studies with respect to gene therapy, transgenics were performed by ICAR-CIFA group of scientists (Barman et.al., 2010; Mohapatra et.al., 2010). In *Labeo rohita* (rohu) myosin light polypeptide chain 2 (mylz 2) promoter which is 1.2 kb were isolated and it can be induced to express in skeletal muscle (Mohanta et. al., 2014; Barman et. al., 2015)

Conclusion

Transgenic fish technology helps in increasing growth rate among food fishes and ornamental fishes. Genetic engineering studies enhance resistance to pathogens which drastically reduces dependence on antibiotics. The genetically modified fishes can withstand high salinity and survive in low oxygen and extremely low temperatures. The transgenic fishes act as models for studying human diseases, embroyonic development studies and also help to understand the mechanism of drug action. Some of transgenic fishes are bioreactors for producing therapeutic proteins like human clotting factor VII or specialized foods. The genetically engineered transgenic fish act as biosensors to detect pollutants and provide real time data on water quality. Transgenic ornamental fishes are household pets enhancing asthetic values. There are number of advantages of transgenetically engineered fishes but there is also risk of contamination the wild species if they escape into natural habitats.

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