

“Zebrafish (*Danio rerio*)” is a New Trend in Biomedical Research as a Model Organism

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ABSTRACT

Animals as model organism, the silent sentinels stand watch over the environmental health of the earth. These are non human animal species which can be utilized to understand certain biological processes and to obtain information on which can be provided an insight into the functioning of other organisms. In the middle of the model organisms, the zebra-fish (*Danio rerio*) is one of the best leading model to study developmental biology, cancer, toxicology, drug discovery, and molecular genetics. Furthermore, the zebrafish is increasingly used as a genetic model organism for aquaculture species and in toxicogenomics as well as to generate zebrafish diseases models for application in human bio-medicines. This minute fish is a versatile model organism for many fields of research because of its easy maintenance, breeding and transparent body in the course of early development Gore et al. (2012).

Keywords: Zebrafish, Model Organism, Cancer, Biological Research

INTRODUCTION

The “Zebrafish model” has been widely used in both animal and human health research, and more recently aquaculture too. In spite of rodents being the most widely used research model in the world. In recent decade the use of the Zebrafish (*Danio rerio*) model has exponentially increased among the scientific community.

The use of Zebrafish as a model organism was pioneered at the University of Oregon, USA by George Streisenger in 1970 because it was simpler Over mouse and easy to manipulate genetically But this species as model organism got impetus from the 1990’s when it was used to developed two large genetic mutants, one by Nobel Prize winner Christiane Nusslein - Volhand in Tubingen, Germany, the other by Wolfgang Driever and Mark Fishman in Boston, USA. According to Kanungo et al. (2014) a lot of physiological and genetic similarities are found

between Zebrafish and humans, including the digestive tract, brain, vasculature, musculature and innate immune system Also 70 percent of human disease genes have functional similarities with those of Zebrafish.

Range:

The Zebrafish is native to fresh water habitats in South Asia where it is found in India, Bangladesh, Nepal and Bhutan.

Habitat:

It is typically inhabit moderately flowing to stagnant clear water of quite shallow depth in streams, canals, ditches, ponds, rice paddies etc.

Description:

Zebrafish named for five uniform pigmented, horizontal blue stripes on side of the body, which are reminiscent of a Zebra’s stripes and which extend to the end of the caudal fin Salient distinguishable structures present between male and female. The Zebrafish reach

Taxonomic Rank	Classification
Kingdom	Animalia
Phylum	Teleostei
Sub-Class	Actinopterygii
Order	Cypriniformes
Family	Cyprinidae
Genus	Danio
Species	Danio rerio

up 4 to 5 cm in length.

Reproduction:

Female spawns every 2 to 3 days, breed all year round external fertilization

Feeding:

Omnivorous

Zebrafish As A Biomedical model Why replace mammal model?

- The mammal models (rodents) can be easily stressed, so some human behaviors cannot be fully explored under these conditions.
- Large space for creation.
- Collecting mouse embryos requires sacrificing the female.

Salient features of Zebrafish as a biomedical model:

- Optically translucent embryos.
- Rapid hatching of eggs.
- The adult fish attains sexual maturity very quickly, with a generation time of about 10 weeks.
- They are cheaper to maintain than mice.
- Amenable for molecular and genetic analysis.
- Zebrafish have the unique ability to repair heart muscle.
- They share 70% of genes with humans. Kalueff_et_al. (2014)

3. Methods and Techniques Used in Zebrafish Research

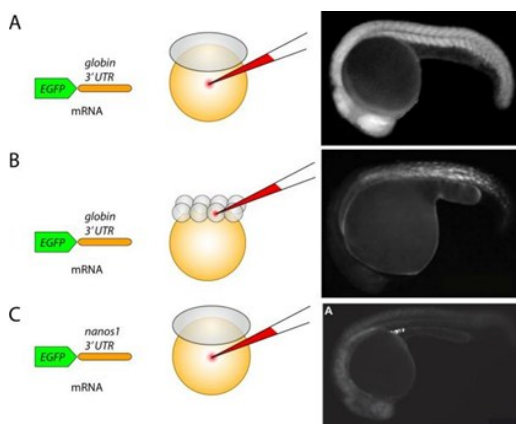


Figure 1: Injection into zebrafish embryos at the early stages of development.

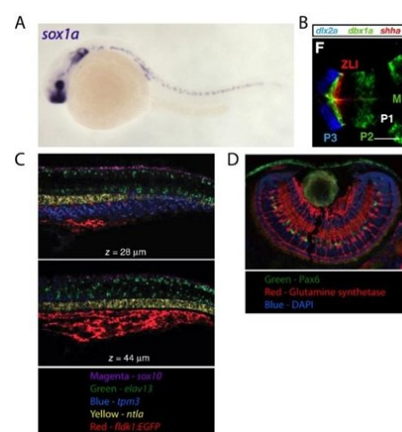


Figure 2: Gene expression and protein visualization.

Use of Zebrafish in developmental biology:

The zebrafish model has been used to see the development of various systems or processes as follows

a) Development of the ENS:

This model species has been made to study the development of the enteric nervous system (ENS) in recent time Guyon et al. (2007). Like all Other vertebrates, the enteric nervous system of Zebrafish is also derived from neural crest, but differs potentially from amniotes where in the enteric nervous system is derived from both the vagal and sacral crests, while in the Case of Zebrafish, it is derived from the vagal crest only.

b) Angiogenesis:

The Zebrafish model has been widely used in the study of angiogenesis and regeneration. Angio- genesis is the process through which new blood vessels original from preexisting Vascular structure which is essential to maintain healthy physiological and pathological conditions.

The Zebrafish exhibits remarkable capacity of regeneration even in adult stages. Due to its simple and fine architecture and relative transparency Caudal fin especially provide an ideal tissue for vascular regeneration studies

The zebrafish as an alternative model for angiogenesis and regeneration studies provide the rele- vance of in vivo assays with simplicity and Versatility of in vitro assays.

4. Zebrafish as a cancer model system

Clinically and pathologically this dreaded disease is found almost exclusively in all vertebrates, from fish to humans. Being a vertebrate the Zebeafish is an ideal model to study cancer, though humans and fishes are separated from their common ancestry but biology of the cancer in both groups of organism is not different. Briggs (2002)

4.1 Carcinogen effects and development of cancer

Fishes are exposed to numerous waterborne carcinogens in the wild which lead to the growth of a variety of benign as

well as malignant tumors in teleosts, and these tumors have similar histology as in humans. Selecting Zebrafish for modeling Cancer studies has many advantages. Extremely conserved cancer pathway can be screened genetically using Zebrafish. Essentially cancer is a disease of adults, but through mutagenesis screens, cell cycle phenotype could be examined in quickly developing transparent embryos of the Zebrafish.

Tumorigeneris

Tumorigeneris is a multi steps process induced by chemical carcinogen, with cumulation of both epi- genetic aberrations and mutations in regulatory regions of genes and disorder of signaling pathways. A variety of fishes to put into action as model to study tumors induced by environmental carcinogen. Among all the Zebrafish proved best for studying embryogenesis, organogenesis and impact of envi- ronmental carcinogen for the growth of cancer. chemically induced tumors in Zebrafish and humans are histopathologically alike, and orthologous oncogenes and tumor suppressor genes (TSGs) have been recognized in fishes and humans. Weinstein (2002) Hepatic gene expression in humans and Zebrafish have has disclosed conservation of gene expression profiles at different stage of tumor aggressiveness between these two phylogenetically outlying species.

Xenotransplantation

It represents another new way to induce tumor in Zebrafish. The significant feature of Xenotransplanta- tion is that tumor cells can be stained or marked using fluorescent stain that differentiate transplanted cells from normal Cells and helps in clear observation of developmental process of the tumor. A number of other types of tumor, such as pancreatic cancer, lung cancer, ovarian carcinoma, breast cancer, prostate cancer, retinoblastoma leukemia etc, have also been transplanted in the Zebrafish.

Skin cancer

Zebrafish is a influential in vivo tool to study pathologies and treatment for skin cancer

(melanoma and SCC). It can be used to study melanoma development, progression, drug screening, and treatment. The Zebrafish model has been utilized recently to recognize the Key molecules which are responsible for the development of cutaneous squamous cell carcinoma (CSCC) and head and neck squamous cell carcinoma well as for SCC target therapies. Bootorabi et al. (2017)

Tumor metastasis

Metastasis, is a multi step and complex process in which tumor cell impale in the vascular system and spread deep in parenchymatous tissues. Because of many noteworthy disadvantages in the previous studies using in Vivo mouse model, the metastasis process can not be abstracted probably, but Zebrafish cancer model has overpower the drawback of previous models and has shown extraordinary strengths.

Toxicology and drug discovery

Because of numerous advantages, the Zebrafish has recently appeared as a prominent model for toxicological studies and drug discovery. The effects of drugs on growth and development can be investigate visually through length and shape of the Zebrafish body as well as morphology of internal organs such as the liver, brain, cardiovascular system, intestine, pancreas, kidney, notochord etc.

Zebrafish embryos are rused as predictive model to assess the toxicity in mammals. The lethal concentration (LC50) of divergent chemicals has been determined in embryos of Zebrafish has been compared with the mammalian LC50 and it has been found that median lethal dose of Zebrafish is lower than mammals. The effects of drugs on specific organs have also been studied, and it has been found that organ toxicity is similar in both Zebrafish and mammals. The drugs that utilized to evaluate the organ toxicity were gentamicin, cisplatin, quinine, vinblastine, doxorubicin, neomycin, cyclosporin A, caffeine etc. Pelegri (2002)

Human disease and Zebrafish

Most of the tissues and organs found in humans and Zebrafish are the identical except lungs and prostate and mammary glands. The cloning of mutated genes screened for specific phenotypes in Zebrafish has resemblance in humans and thus serves as model for human disease and to study underlying mechanisms the first human disease identified using Zebrafish was a blood disorder relate to specific defect in hemoglobin production through ALAS2 mutated gene. Many other mutants which show phenotypic comparability to human disease have been screened and identified. These include neurological disorders, hematological disorder, Cardiovascular diseases, muscle disease and Cancers, Parkinson's disease, anxiety and post-traumatic stress disorder. Guyon et al. (2007)

Zebrafish as a model organism for aquaculture species

Among the different fish species of interest to aquaculture, Zebrafish is genetically more tractable. The Zebrafish model is used commercially in a lot of areas of aquaculture such as in the identification of genes involved in the development of the muscles, bones and fats, the metabolism of nutrients, disease and stress physiology and also behavioural traits. Bailone et al. (2020)

Future Prospects (Conclusion)

- Zebrafish have the rare ability to repair their own tissue, including spinal cord, retina, and heart—understanding this could lead to the development of new human treatments.
- Zebrafish are becoming an increasingly successful model to understand Alzheimer's disease, with future scope for investigation in neurodevelopment and neurodegeneration, which promises exciting research opportunities in the future.
- The speed and innovation advantages of zebrafish offer future opportunities to expand the positive impact of our discipline and help to meet the challenges facing 21st-century societies.

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