Int. J. Sci. R. Tech., 2025 2(5)
A Multidisciplinary peer-reviewed Journal
www.ijsrtjournal.com [ISSN: 2394-7063]

Original Article

Zebrafish Model in Pharmaceutical Research: A Review

Vaibhavi Pagar[1], Gaurav Kasar*[1], Dipti Chavan[1], Dr. Chandrashekhar Patil[1], Dr. Sunil Mahajan[2]

[1]*Department of Pharmacology, Divine College of Pharmacy, Satana (Nashik), India*
[2]*Department of Pharmaceutical Chemistry, Divine College of Pharmacy, Satana (Nashik), India*

ABSTRACT

Zebrafish (Danio rerio) has emerged as a powerful and versatile model organism in pharmaceutical research due to its unique biological and genetic attributes. With approximately 70% genetic similarity to humans and functional conservation of disease-related genes, zebrafish serve as an efficient platform for studying human diseases, drug screening, and toxicity assessment. Their small size, rapid development, and high fecundity make them ideal for large-scale in vivo experiments, particularly in developmental biology, pharmacology, and toxicology. Transparency of embryos allows real-time imaging of physiological and pathological processes, enhancing their applicability in high-throughput drug screening. Zebrafish models have been instrumental in advancing our understanding of neurological disorders, cardiovascular diseases, metabolic syndromes and cancer. However, advancements in genetic engineering, imaging technologies, and automation are addressing these challenges. This review provides a comprehensive overview of zebrafish applications in pharmaceutical research, discussing their advantages, limitations, and future prospects in drug development and translational medicine.

Keywords: Zebrafish model, Research, Genetic, Pharmacology

INTRODUCTION

Zebrafish (*Danio rerio*), a small freshwater fish native to South Asia, has gained significant attention as a powerful model organism in biomedical and pharmaceutical research. Due to its unique biological and genetic characteristics, zebrafish has become an essential tool for studying human diseases, drug screening, and toxicity assessment [1]. Compared to traditional mammalian models such as mice and rats, zebrafish offer several advantages, including their ease of maintenance, rapid development, and high-throughput screening potential. These attributes make zebrafish a cost-effective and efficient alternative for studying various aspects of human health and disease [2,3]. Their use has expanded in recent years, particularly in developmental biology, toxicology, pharmacology, and genetic research. Biologically, zebrafish possess several key characteristics that make them ideal for laboratory studies. They are small in size, measuring about 2.5–5 cm in length, and have a relatively short lifespan of around 2–3 years. Their external fertilization and transparent embryos allow for real-time observation of early developmental processes, providing a unique advantage over mammalian models, where embryonic development occurs internally. One of the most notable features of zebrafish is their rapid development, with major organ systems forming within the first 24–48 hours post-fertilization [4]. This makes them highly suitable for studying embryogenesis, organogenesis, and genetic modifications in real time. Additionally, zebrafish exhibit a high reproductive rate, with a single female capable of laying hundreds of eggs per week. This large number of offspring facilitates large-scale experiments, making zebrafish particularly useful for genetic studies and drug screening applications [5,6]. Genetically, zebrafish share remarkable similarities with humans, making them highly relevant for translational research. Approximately 70% of zebrafish genes are homologous to human genes, and about 84% of human disease-related genes have a corresponding zebrafish counterpart. This genetic conservation allows researchers to study various human diseases in zebrafish models, including neurological disorders, cardiovascular diseases, metabolic syndromes, and cancers. Moreover,

Relevant conflicts of interest/financial disclosures: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

zebrafish possess well-developed organ systems that closely resemble those of humans, including the nervous, cardiovascular, immune, and digestive systems. Their functional drug targets, such as ion channels, enzymes, and receptors, are also conserved, making zebrafish an effective model for pharmacological studies [7-9]. Due to these genetic and physiological similarities, zebrafish have become an invaluable tool for studying gene function, disease mechanisms, and drug responses in a whole-organism context. The zebrafish has emerged as a powerful model organism in pharmaceutical research due to its unique biological and physiological characteristics. Initially used in developmental biology, zebrafish are now widely recognized for their utility in pharmacology, toxicology, and disease modeling. Their small size, rapid life cycle, and high fecundity make them particularly attractive for large-scale studies, including high-throughput screening of pharmaceutical compounds. Additionally, their external fertilization and optical transparency during embryonic and larval stages enable real-time imaging and in vivo analysis of physiological and pathological processes. One of the most significant advantages of zebrafish in pharmaceutical research is their genetic homology with humans. Approximately 70% of zebrafish genes are shared with humans, and around 84% of human disease-related genes have counterparts in zebrafish. This genetic similarity allows researchers to develop zebrafish models for a variety of human diseases, including cancer, neurodegenerative disorders, cardiovascular conditions, and metabolic diseases. The advent of advanced genetic manipulation techniques, such as CRISPR-Cas9 genome editing and transgenic technologies, has further strengthened the role of zebrafish as a versatile model for studying gene functions and drug responses [10-13].

Figure 1: Zebrafish

Zebrafish also offer significant advantages in toxicity testing and pharmacokinetics. Their embryos and larvae are highly sensitive to toxicants, allowing researchers to assess the safety profile of drugs in a cost-effective and ethically favorable manner. Because zebrafish can absorb small molecules directly from water, they provide an efficient and reliable system for studying drug absorption, distribution, metabolism, and excretion (ADME) [14]. The presence of cytochrome P450 enzymes in zebrafish further facilitates pharmacokinetic studies that are comparable to mammalian models. Furthermore, zebrafish models contribute to personalized medicine by enabling the development of patient-derived xenograft (PDX) models, where human tumor cells are implanted into zebrafish embryos to assess drug sensitivity. This approach allows for rapid and individualized drug testing, offering potential breakthroughs in precision medicine. Despite their many advantages, zebrafish models also have limitations. Some physiological differences, such as the absence of lungs and adaptive immunity, can restrict their use in certain disease studies. Additionally, their small size limits the amount of blood and tissue available for biochemical analysis, posing challenges for pharmacokinetic studies that require large sample volumes. Nonetheless, continuous advancements in zebrafish research methodologies, including automation, imaging technologies, and computational modeling, are addressing these limitations and expanding their applicability in pharmaceutical sciences. This review explores the role of zebrafish in various aspects of pharmaceutical research, including drug discovery, toxicity testing, disease modeling, and pharmacokinetics. It also highlights the advantages

and challenges associated with zebrafish models and discusses future perspectives on their integration into drug development and translational medicine [15].

Advantages of Zebrafish Model in Pharmaceutical Research

- **Genetic Similarity to Humans**: Zebrafish share approximately 70% of human genes, and about 84% of genes associated with human diseases have a counterpart in zebrafish. This genetic homology allows researchers to study genetic diseases, drug interactions, and gene functions in a system that closely resembles human physiology.

- **Transparency and Imaging**: The optical transparency of zebrafish embryos and larvae facilitates real-time imaging of organ development and drug effects using advanced fluorescence microscopy. This feature is particularly useful in studying neurobiology, cardiovascular systems, and cancer progression.

- **High Reproductive Rate**: A single pair of zebrafish can produce hundreds of embryos weekly. This rapid reproduction allows for large-scale studies and high-throughput screening of drug candidates, significantly reducing the time required for preclinical testing.

- **Cost-effectiveness**: Compared to rodent models, zebrafish require significantly lower costs for housing, feeding, and experimental setups. This economic advantage makes them a preferred model for large-scale pharmaceutical screenings.

- **Ethical Considerations**: Zebrafish, being lower vertebrates, are subject to fewer ethical constraints than mammals. This enables researchers to conduct large-scale studies with fewer regulatory hurdles, particularly in early-stage drug development [16].

Drug Screening in Neurological Disorders

Zebrafish serve as a powerful model for studying neurological disorders due to their evolutionary similarity to the human nervous system. Their well-developed neuroendocrine system, along with homologous brain structures including the diencephalon, telencephalon, cerebellum, and spinal cord makes them valuable for neurodevelopmental studies and drug discovery.

Zebrafish Models in Depression Research

Developing effective therapeutics for neurological disorders remains a challenge, with depression being one of the most prevalent and debilitating conditions, affecting over 20% of the global population. Environmental stress and neurochemical imbalances in zebrafish parallel those seen in humans. For instance, the grs357 zebrafish mutant, which carries a glucocorticoid receptor mutation, exhibits altered corticosteroid feedback, transcriptional dysregulation, and behavioral abnormalities such as reduced locomotion and heightened startle responses. Treatment with fluoxetine and diazepam in both wild-type and mutant zebrafish effectively blocked stress-induced upregulation of the mineralocorticoid receptor (MR) and serotonin transporter (Serta), highlighting the potential of zebrafish in antidepressant drug screening. These findings underscore the conserved relationship between the hypothalamic–pituitary–adrenal (HPA) axis and affective disorders across vertebrates, supporting zebrafish as a high-throughput model for antidepressant discovery.

Zebrafish Models in Parkinson's Disease Research

Zebrafish models have provided critical insights into the genetics and pathophysiology of Parkinson's disease (PD), a progressive neurodegenerative disorder characterized by dopaminergic neuron loss and Lewy body formation. The zebrafish nervous system lacks a distinct midbrain dopaminergic region like mammals but possesses a homologous dopaminergic system in the posterior tuberculum of the diencephalon. Exposure to the neurotoxin MPTP (1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine) induces Parkinson's-like symptoms in zebrafish by selectively depleting dopaminergic neurons. Studies have shown that deprenyl, a monoamine oxidase B (MAO-B) inhibitor, counteracts MPTP-induced neurotoxicity by preventing its conversion to the toxic metabolite MPP+. Similarly, co-treatment with nomifensine, a dopamine transporter (DAT) inhibitor, protects against dopaminergic neuron loss. Proteomic analysis of MPTP-treated zebrafish models has

identified key differentially expressed proteins, including NEFL, MUNC13-1, NAV2, and GAPVD1, which are downregulated in the diseased state. Another study demonstrated that 6-hydroxydopamine (6-OHDA) exposure leads to locomotor deficits and dopaminergic cell loss, which can be reversed with vitamin E, sinemet, or minocycline. These findings validate zebrafish as a reliable in vivo model for Parkinson's disease research.

Zebrafish Models in Huntington's Disease Research

Huntington's disease (HD) is an autosomal-dominant neurodegenerative disorder caused by mutations in the HTT gene, leading to the accumulation of toxic huntingtin (HTT) protein aggregates. Since complete HTT gene deletion in zebrafish results in embryonic lethality, morphant zebrafish models provide an alternative platform for studying HD pathology. Treatment with brain-derived neurotrophic factor (BDNF) in HTT knockdown zebrafish has been shown to significantly improve disease phenotypes, highlighting its therapeutic potential. To model HD in adult vertebrates, researchers have employed quinolinic acid (QA) injections in the striatum of rodents to induce neurotoxicity. Similar lesioning in the zebrafish telencephalon has been found to stimulate robust neuroregeneration, with neural stem cells integrating new neurons over long distances. Furthermore, a study evaluating the neuroprotective effects of Centella asiatica hydroalcoholic extract in zebrafish with 3-nitropropionic acid-induced HD demonstrated its antioxidant and anti-inflammatory properties, offering promising therapeutic implications [17-21].

Applications of Zebrafish in Pharmaceutical Research

Drug Discovery and Screening Zebrafish have become a crucial model for high-throughput drug screening due to their ability to absorb small molecules from water. Drug candidates can be directly added to the aquatic environment, and their effects can be observed in vivo. Several drugs, including anti-cancer agents, antibiotics, and neuroactive compounds, have been identified and validated using zebrafish models. The use of automated imaging and behavioral assays has further enhanced the drug discovery process by enabling rapid assessment of drug efficacy and toxicity [22].

Toxicity Testing Toxicological assessments in zebrafish provide critical insights into the safety profile of pharmaceutical compounds. Zebrafish embryos and larvae are highly sensitive to toxicants, making them an ideal model for detecting cardiotoxicity, hepatotoxicity, neurotoxicity, and teratogenicity. The model allows researchers to study dose-response relationships and identify adverse effects at early developmental stages. Notably, zebrafish-based toxicity studies have been successfully used in predicting human toxicity, making them a valuable alternative to traditional mammalian models [23].

Disease Modeling The genetic and physiological similarities between zebrafish and humans enable researchers to model various human diseases. Genetic modifications using CRISPR-Cas9 and transgenic techniques have led to the development of zebrafish models for diseases such as:

- **Cancer**: Zebrafish models have been used to study melanoma, leukemia, and breast cancer, providing valuable insights into tumor progression, metastasis, and drug resistance mechanisms.
- **Neurodegenerative Disorders**: Models for Alzheimer's disease, Parkinson's disease, and epilepsy have been developed to investigate disease pathology and screen potential therapeutics.
- **Cardiovascular Diseases**: Zebrafish heart development closely resembles that of humans, allowing for the study of congenital heart defects, arrhythmias, and drug-induced cardiotoxicity.
- **Metabolic Disorders**: Diabetes and obesity models have been established to explore metabolic pathways and assess anti-diabetic drugs.

Pharmacokinetics and Pharmacodynamics Zebrafish provide valuable insights into the absorption, distribution, metabolism, and excretion (ADME) of drugs. The presence of cytochrome P450

enzymes, which play a critical role in drug metabolism, enables pharmacokinetic studies comparable to those in humans. Additionally, fluorescent and luminescent reporter assays allow real-time monitoring of drug distribution and target engagement within live zebrafish embryos [24].

Limitations of the Zebrafish Model

- **Physiological Differences**: While zebrafish share many genetic similarities with humans, some physiological systems, such as lungs and adaptive immunity, differ significantly. This can limit their applicability in certain disease studies.
- **Limited Blood Volume**: The small size of zebrafish limits the amount of blood that can be collected, posing challenges for pharmacokinetic and pharmacodynamic studies requiring large sample volumes.
- **Differences in Drug Metabolism**: Some metabolic pathways in zebrafish differ from humans, leading to variations in drug responses. This necessitates validation in mammalian models before clinical translation [25,26].

FUTURE PERSPECTIVES

Advancements in genetic engineering, imaging technologies, and automated screening techniques are expected to enhance the applicability of zebrafish in pharmaceutical research. Emerging technologies such as single-cell RNA sequencing, optogenetics, and CRISPR-based gene editing are improving the precision and reliability of zebrafish models [27]. Furthermore, integration with computational modeling and organ-on-a-chip technologies holds the potential to bridge the gap between preclinical studies and human trials. Future research efforts should focus on refining zebrafish models to improve their predictive accuracy for human drug responses.

CONCLUSION

The zebrafish model has revolutionized pharmaceutical research by offering a cost-effective, high-throughput, and genetically relevant platform for studying human diseases and evaluating drug candidates. Its genetic similarity to humans, coupled with its rapid development and transparency, has enabled significant advancements in drug discovery, toxicity testing, and disease modeling. Zebrafish-based research has contributed to understanding complex disorders such as Parkinson's disease, Huntington's disease, and cancer while also facilitating personalized medicine through patient-derived xenograft models. However, challenges such as physiological differences and limited sample availability necessitate further optimization of zebrafish methodologies. Emerging technologies, including CRISPR-Cas9 gene editing, single-cell RNA sequencing, and computational modeling, are enhancing the precision and applicability of zebrafish models. As these advancements continue, zebrafish are expected to play an increasingly vital role in bridging the gap between preclinical studies and human clinical trials, making them an indispensable tool in modern pharmaceutical research.

REFERENCE

1. Cong L et al. 2013 Multiplex genome engineeringusing CRISPR/Cas9, systems. Science 339, 819–823. (doi:10.1126/science.1231143)
2. Mali P, Yang L, Esvelt KM, Aach J, Guell M, DiCarloJE, Church GM. 2013 RNA-guided human genomeengineering via Cas9. Science 339, 823–826. (doi:10.1126/science.1232033)
3. Murphey RD, Zon LI. 2006 Small moleculescreening in the zebrafish. Methods 39,255–261. (doi: 10.1016/j.ymeth.2005.09.019)
4. Howe K et al. 2013 The zebrafish referencegenome sequence and its relationship tothe human genome. Nature 496, 498–503. (doi:10.1038/nature12111)
5. Thayumanavan G et al. 2022 Silibinin andnaringenin against bisphenol A-inducedneurotoxicity in zebrafish model-potential flavonoidmolecules for new drug design, development, andtherapy for neurological disorders. Molecules 27,2572. (doi:10.3390/molecules27082572)
6. Raldúa D, Piña B. 2014 In vivo zebrafish assaysfor analysing drug toxicity. Expert Opin. DrugMetab. Toxicol. 10, 685–697. (doi:10.1517/17425255.2014.896339)
7. Oprişoreanu AM, Smith HL, Krix S, Chaytow H, Carragher NO, Gillingwater TH, Becker CG, Becker T. 021 Automated in vivo drug screen in

zebrafishidentifies synapse-stabilising drugs with relevanceto spinal muscular atrophy. Dis. Model Mech. 14, dmm047761. (doi:10.1242/dmm.047761

8. White DT et al. 2016 ARQiv-HTS, a versatile whole-organism screening platform enabling in vivo drugdiscovery at high-throughput rates. Nat. Protoc. 11,2432–2453. (doi:10.1038/nprot.2016.142)
9. Lubin A, Otterstrom J, Hoade Y, Bjedov I, Stead E,Whelan M, Gestri G, Paran Y, Payne E. 2021 Aversatile, automated and high-throughput drugscreening platform for zebrafish embryos. Biol. Open10, bio058513. (doi:10.1242/bio.058513)
10. Chan J, Bayliss PE, Wood JM, Roberts TM. 2002Dissection of angiogenic signaling in zebrafish usinga chemical genetic approach. Cancer Cell. 1,257–267. (doi:10.1016/S1535-6108(02)00042-9)
11. Shinya M, Koshida MS, Sawada A, Kuroiwa A, Takeda H.2001 Fgf signalling through MAPK cascade is requiredfor development of the subpallial telencephalon inzebrafish embryos. Development 128, 4153–4164. (doi:10.1242/dev.128.21.4153)
12. Taylor KL, Grant NJ, Temperley ND, Patton EE. 2010Small molecule screening in zebrafish: an in vivoapproach to identifying new chemical tools anddrug leads. Cell Commun. Signal 8, 11. (doi:10.1186/1478-811X-8-11)
13. MacRae CA, Peterson RT. 2015 Zebrafish as tools fordrug discovery. Nat. Rev. Drug Discov. 14, 721–731. (doi:10.1038/nrd4627)
14. Rennekamp AJ, Peterson RT. 2015 15 years ofzebrafish chemical screening. Curr. Opin. Chem. Biol.24, 58–70. (doi: 10.1016/j.cbpa.2014.10.025)
15. Ito T, Ando H, Suzuki T, Ogura T, Hotta K, ImamuraY, Yamaguchi Y, Handa H 2010 Identification of aprimary target of thalidomide teratogenicity. Science327, 1345–1350. (doi:10.1126/science.1177319)
16. Patton EE, Zon LI, Langenau DM. 2021 Zebrafishdisease models in drug discovery: from preclinicalmodelling to clinical trials. Nat. Rev. Drug Discov.20, 611–628. (doi:10.1038/s41573-021-00210-8)
17. 1MacRae CA, Peterson RT. 2003 Zebrafish-based smallmolecule discovery. Chem. Biol. 10, 901–908. (doi: 10.1016/j.chembiol.2003.10.003)
18. Zon LI, Peterson RT. 2005 In vivo drug discovery inthe zebrafish. Nat. Rev. Drug Discov. 4, 35–44. (doi:10.1038/nrd1606)
19. Richter S, Schulze U, Tomançak P, Oates AC. 2017 Smallmolecule screen in embryonic zebrafish using modularvariations to target segmentation. Nat. Commun. 8,1901. (doi:10.1038/s41467-017-01469-5)
20. Wiley DS, Redfield SE, Zon LI. 2017 Chemicalscreening in zebrafish for novel biological andtherapeutic discovery. Methods Cell Biol. 138,651–679. (doi: 10.1016/bs.mcb.2016.10.004)
21. Swanhart LM, Cosentino CC, Diep CQ, Davidson AJ,de Caestecker M, Hukriede NA. 2011 Zebrafishkidney development: basic science to translationalresearch. Birth Defects Res. C Embryo Today 93,141–156. (doi:10.1002/bdrc.20209)
22. Lipinski CA, Lombardo F, Dominy BW, Feeney PJ.2001 Experimental and computational approachesto estimate solubility and permeability in drugdiscovery and development settings. Adv. DrugDeliv. Rev. 46, 3–26. (doi:10.1016/S0169-409X (00)00129-0)
23. Lipinski CA. 2004 Lead- and drug-like compounds: the rule-of-five revolution. Drug Disc. Today 1,337–341. (doi: 10.1016/j.ddtec.2004.11.007)
24. Long K, Kostman SJ, Christian Fernandez C, BurnettJC, Donna M, Huryn DM. 2019 Do zebrafish obeylipinski rules? ACS Med. Chem. Lett. 10, 1002–1006. (doi:10.1021/acsmedchemlett.9b00063)
25. Kalueff AV, Stewart AM, Gerlai R. 2014 Zebrafish asan emerging model for studying complex braindisorders. Trends Pharmacol. Sci. 35, 63–75. (doi: 10.1016/j.tips.2013.12.002)
26. Kessler RC, Berglund P, Demler O, Jin R, Merikangas KR, Walters EE. 2005 Lifetime prevalence and age-of-onsetdistributions of DSM-IV disorders in the National Comorbidity Survey Replication. Arch. Gen. Psychiatry62, 593–602. (doi:10.1001/archpsyc.62.6.593)
27. Ziv L, Muto A, Schoonheim PJ, Meijsing SH, StrasserD, Ingraham HA, Schaaf MJ, Yamamoto

KR, Baier H.2013 An affective disorder in zebrafish withmutation of the glucocorticoid receptor. Mo. Psychiatry 18, 681–691. (doi:10.1038/mp.2012.64).

HOW TO CITE: Vaibhavi Pagar, Gaurav Kasar*, Dipti Chavan, Dr. Chandrashekhar Patil, Dr. Sunil Mahajan, Zebrafish Model in Pharmaceutical Research: A Review, Int. J. Sci. R. Tech., 2025, 2 (5), 535-541. https://doi.org/10.5281/zenodo.15507701