ISSN: 2694-166X
Scientific Journal
Biology & Life Sciences
DOI: 10.33552/SJBLS.2024.03.000573
Iris Publishers

Mini Review

Navigating Disease with Zebrafish: A Promising Model for Biomedical Research

Youchao Chen*

Warwick Medical School, University of Warwick, Coventry, CV4 7AL, United Kingdom

***Corresponding author:** Youchao Chen, Warwick Medical School, University of Warwick, Coventry, CV4 7AL, United Kingdom

Received Date: July 07, 2024

Published Date: August 05, 2024

Introduction

Effectively studying human diseases necessitates the use of appropriate tools, with one of the most essential being an accurate animal model that faithfully reproduces the human condition. Zebrafish (Danio Rerio) has become a pivotal model organism in biomedical research, leveraging its unique biological features and genetic similarities to humans. Zebrafish possess similar organs to mammals [1], encompassing the brain, heart, liver, spleen, pancreas, gallbladder, intestines, kidneys, testes, and ovaries (Figure 1). The genome of zebrafish has been sequenced and annotated. Around 70% genetic sequence similarity with humans. Within this genetic similarity, 84% aligns with known human-associated diseases [2]. This high level of homology to humans offers a valuable platform for studying disease-related genes and mechanisms, accelerating insights into human diseases. Their rapid development, optical transparency, high fecundity, and scalability contribute to efficient experimentation, making zebrafish a robust model for disease studies (Figure 1).

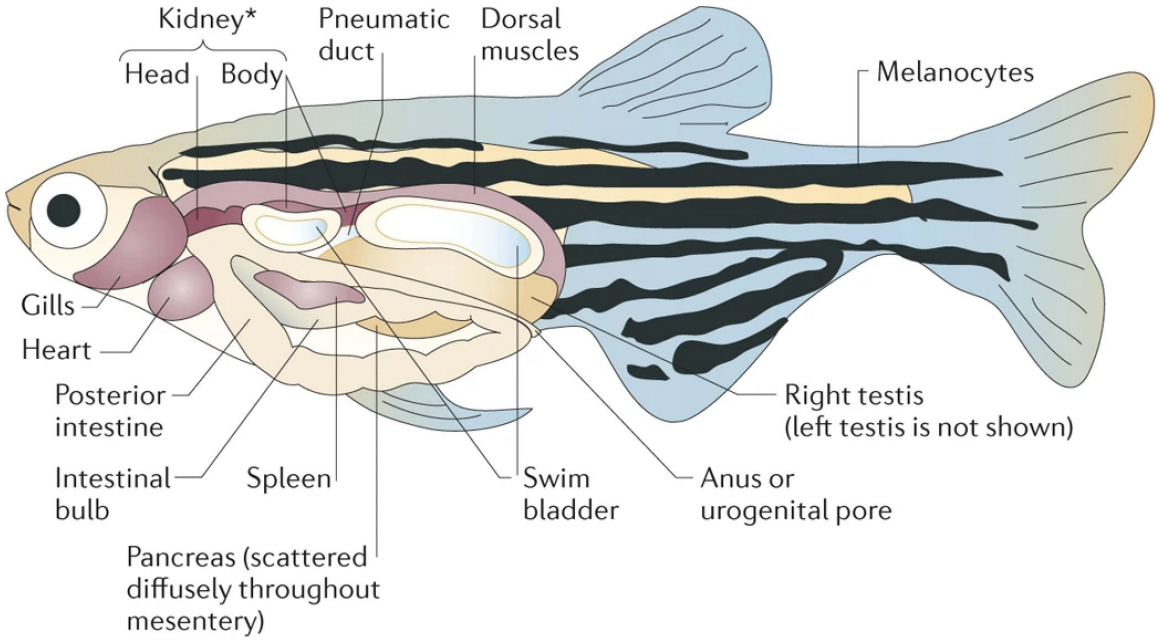

Figure 1: Zebrafish anatomy. An adult zebrafish is shown with the anatomical structures labelled. Zebrafish share most of their organs with mammalian counterparts, including the brain, heart, liver, spleen, pancreas, gallbladder, intestines, kidney, testis and ovaries. Taken from [1].

CRISPR/Cas9 technology in zebrafish genome editing

The CRISPR-Cas9 nuclease system has emerged as a highly effective and widely adopted gene editing technology in zebrafish models [3]. Its distinct advantages, including a straightforward design process and rapid reagent synthesis, set it apart from other methods like zinc finger nucleases (ZFNs) and transcription activator-like effector nucleases (TALENs) [4]. While morpholinos (MOs) have traditionally been used for transient gene knockdown, CRISPR-Cas9 offers the advantage of producing permanent genetic modifications with relatively low off-target effects [3]. This approach effectively utilizes zebrafish as a model for studying genetic diseases.

In the CRISPR/Cas9 system (Figure 2), a guide RNA binds to a 20-nucleotide DNA sequence located immediately before an NGG DNA motif known as the protospacer-associated motif (PAM). This binding results in a double-strand break (DSB) occurring 3 base pairs upstream of the NGG. The cellular DNA repair machinery then utilizes the DSBs as substrates, catalysing either non-homologous end joining (NHEJ) or homology-directed repair (HDR) [3]. The enzyme, guided by an RNA molecule with a matching sequence to the cleavage site, creates specific DSBs marked by PAM sequences. RNA-guided Cas9 activity induces precise double-stranded DNA breaks, subsequently repaired through either NHEJ or HDR. In homologous recombination, the introduction of donor DNA facilitates the insertion of new sequence information at the break site [3].

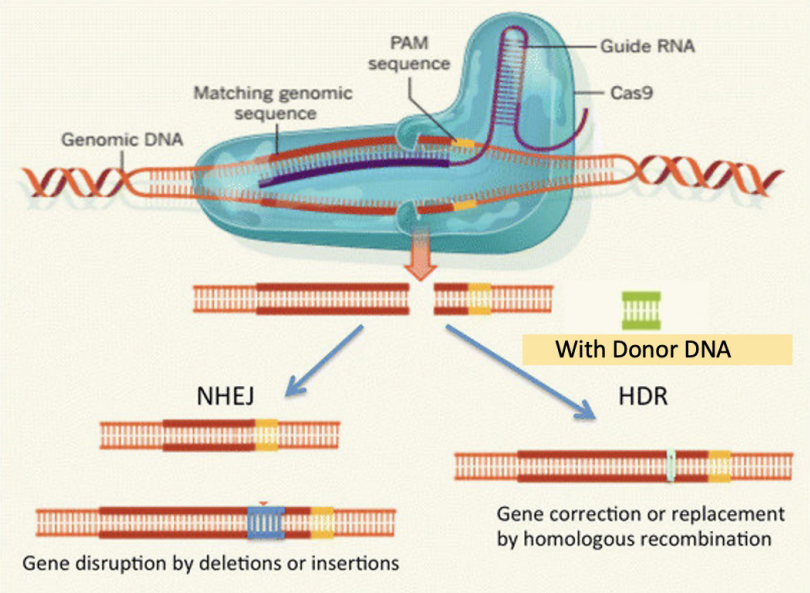

Figure 2: In the CRISPR/Cas9 system, a guide RNA hybridizes a 20-nt DNA sequence immediately preceding an NGG DNA motif (PAM), resulting in a double-strand break (DSB) 3 bp upstream of the NGG. The double-stranded DNA breaks become substrates for endogenous cellular DNA repair machinery that catalyzes nonhomologous end joining (NHEJ) or homology-directed repair (HDR). The enzyme is guided to the target DNA by an RNA molecule that contains a sequence that matches the sequence to be cleaved, which is demarcated by PAM sequences. RNA-guided Cas9 activity creates site-specific double-stranded DNA breaks, which are then repaired by either non-homologous end joining or homologous recombination. During homologous recombination, the addition of donor DNA enables new sequence information to be inserted at the break site. Modified from [3].

An up to date system utilizes a ribonucleoprotein (RNP) complex composed of guide RNA (gRNA) and the Cas9 nuclease to generate F0 mutants [5]. The gRNA comprises a target-specific CRISPR RNA (crRNA) and a universal trans-activating RNA (tracrRNA). Specifically designed to be complementary to the target DNA site, the crRNA forms a complex with the unique target site, guiding the Cas9 nuclease to the intended locus. Upon reaching the target site, Cas9 induces DSB, initiating the NHEJ repair mechanism. The error-prone nature of NHEJ leads to small insertions or deletions between the broken DNA strands, resulting in efficient mutagenesis [3,6]. This permanent and precise gene editing capability positions CRISPR-Cas9 as a valuable tool for advancing genetic studies and functional genomics in zebrafish research.

Conclusion

This mini review has underscored the invaluable role of zebrafish as a versatile and powerful model organism in biomedical research, especially in studying genetic diseases. Zebrafish offer unique advantages, including their rapid development, optical transparency during early stages, and genetic tractability, which collectively enable researchers to efficiently study disease mechanisms and therapeutic interventions in vivo. Zebrafish models can not only provide insights into disease pathogenesis but also facilitate high-throughput screening of potential drug candidates

and elucidate the underlying molecular pathways involved. Looking forward, ongoing advancements in genome editing technologies, imaging modalities, and high-throughput screening methods promise to further enhance the utility and scope of zebrafish in biomedical research. Collaborative efforts across disciplines will continue to drive innovation and accelerate the translation of findings from zebrafish models into clinical applications, ultimately benefiting human health. Embracing zebrafish as a complementary model system alongside traditional mammalian models holds great promise for advancing our understanding of disease mechanisms and developing effective treatments in the future.

References

1. White R, Rose K, Zon L (2013) Zebrafish cancer: the state of the art and the path forward. Nat Rev Cancer 13(9): 624-636.
2. Choi TY, Choi TI, Lee YR, Choe SK, Kim CH (2021) Zebrafish as an animal model for biomedical research. Exp Mol Med 53(3): 310–317.
3. Charpentier E, Doudna JA (2013) Rewriting a genome. Nature 495(7439): 50–51.
4. Makarova KS, Wolf YI, Iranzo J, Shmakov SA, Alkhnbashi OS, et al. (2020) Evolutionary classification of CRISPR-Cas systems: a burst of class 2 and derived variants. Nat Rev Microbiol 18(2): 67-83.
5. Hoshijima K, Jurynec MJ, Klatt Shaw D, Jacobi AM, Behlke MA, et al. (2019) Highly Efficient CRISPR-Cas9-Based Methods for Generating Deletion Mutations and F0 Embryos that Lack Gene Function in Zebrafish. Dev Cell 51(5): 645-657.e4.
6. Varshney GK, Pei W, LaFave MC, Idol J, Xu L, et al. (2015) High-throughput gene targeting and phenotyping in zebrafish using CRISPR/Cas9. Genome Res 25(7): 1030-1042.