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Zebrafish (*Danio rerio*) – this small animal has made a "big splash" in research

Tiny Heroes of Biological Research

Zebrafish (*Danio rerio*) have transformed biological research, providing insights into many biological processes and complex human diseases

Ewa Stefanik
Karolina Wolny

Department of Animal Developmental Biology, Faculty of Biological Sciences, University of Wrocław

The zebrafish (*Danio rerio*), named for the characteristic blue stripes running along its body, is just centimeters long. While native to South Asia, it can now be found in many home aquariums around the world. However, the zebrafish has gained immense popularity not just among aquarium enthusiasts, but also among scientists from various fields. It is widely used as a model organism, meaning that research on zebrafish can yield findings applicable to a wider range of species. In other words, by studying the genetic mechanisms in zebrafish, we can indirectly gain insights into those operating in humans.

This is possible despite the 400 million years of evolution that separate humans and zebrafish, as their developmental lines diverged long ago. However, when considering the 3.5 billion years of life on Earth, it's clear that even after such a long split, common

Ewa Stefanik, MSc
is a doctoral candidate at the Doctoral College of Biological Sciences at the University of Wrocław. Founder of the popular science project "Zebrafish Adventure" and a board member of the "March for Science" Foundation. Recipient of a Sonata 17 grant.
ewa.stefanik2@uwr.edu.pl

Karolina Wolny, MSc
holds a degree in genetics and experimental biology at the University of Wrocław. Chair of the Student Scientific Circle of Geneticists "Genome Guardians" at the University. She is engaged in science communication and research in the field of skeletal muscle development and physiology.
321274@uwr.edu.pl

biological features have persisted. While there are clear differences at the cellular level, the organs of zebrafish and humans – like the heart, brain, and muscles – function in remarkably similar ways. This is due to their genetic similarities. Comparative studies have shown that up to 80% of zebrafish genes have counterparts in the human genome.

Natural and enhanced traits

Several key traits make the zebrafish an invaluable tool for studying biological processes. They reproduce quickly, allowing researchers to complete experiments in a short amount of time. They also produce large numbers of offspring – up to 300 eggs from a single pair – enabling many experimental repetitions. One of the most useful features of zebrafish is that their eggs are transparent and develop outside the female's body. This transparency allows scientists to observe embryonic development right from the first moments of life. Similarly, the transparent bodies of zebrafish larvae enable researchers to closely study the development of key organs, like the heart and skeletal muscles, in real time. Another benefit is their short life cycle. Within just three months of fertilization, zebrafish reach sexual maturity, allowing researchers to analyze multiple generations in a relatively short time. Additionally, their small size makes it easier and more cost-effective to maintain large populations in the lab.

Another major advantage of zebrafish is the ease of genetic manipulation within their genome. By creating models of human diseases in zebrafish, scientists can study the functions of specific genes, their roles in metabolic pathways, and the effects of genetic mutations. Zebrafish are a well-established model for studying cancers like melanoma, sarcoma, thyroid cancer, leukemia, and many others. Today, scientists have access to various techniques to create specialized zebrafish strains that further support research. One such strain is called Casper. Typically, the transparency of zebrafish bodies lasts for the first five days of life, after which they start producing pigment, giving them their usual white-blue coloring. However, the Casper strain has been genetically modified to remain transparent throughout its life. This transparency makes it ideal for cancer research, as it allows scientists to monitor tumor growth and metastasis in real time. By introducing fluorescent cancer cells into the zebrafish, researchers can observe the glowing cells through its transparent skin, tracking cancer progression as it happens.

Beyond cancer research

Zebrafish are also proving to be an invaluable model for studying cardiovascular diseases, which remain one of the leading causes of death worldwide. Despite advances in medicine, finding effective ways to prevent and treat heart conditions remains a challenge. In mammals, heart cells lose the ability to divide, so once the heart is damaged, it can't fully regenerate. Zebrafish, however, have an extraordinary ability to regenerate tissues. When damaged, the fish's organs, including the heart, can fully recover their function. By understanding this regeneration process, we may in the future develop better treatments for patients who suffer from heart attacks.

Yet another surprising advantage of zebrafish is how closely their heart's electrophysiology resembles that of humans. The heartrate of mice – a common model for research – is 10 times faster than that of humans, but that of the zebrafish matches the human heartbeat. This makes zebrafish ideal for studying heart rhythm disorders. The transparent bodies of zebrafish larvae and their external development make it possible to observe changes in the heart from the earliest stages of life, which is crucial for researching

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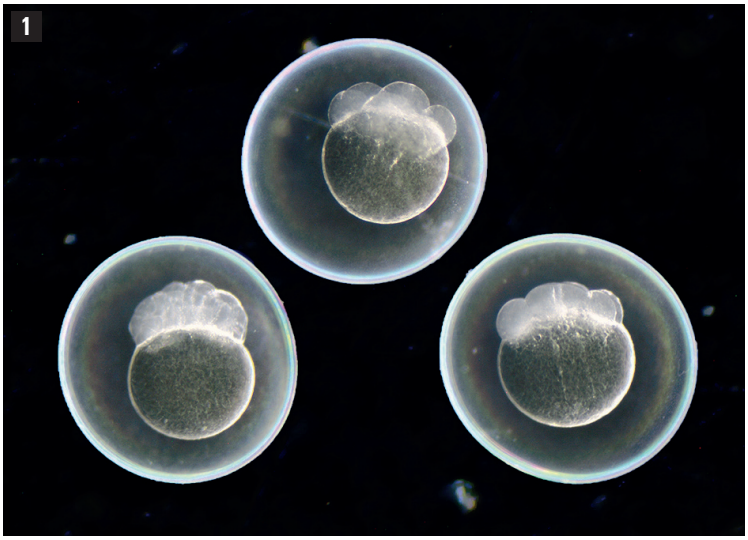
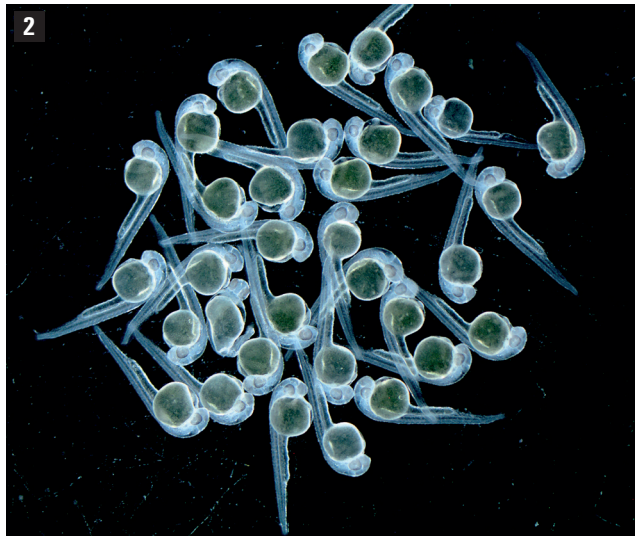
The Casper strain of zebrafish, such as this specimen, remains transparent throughout its life, allowing scientists to peer inside its body





JOANNA NIEDBALSKA-TARNOWSKA (2)

congenital heart defects. Many of these defects arise from developmental issues during embryonic growth. Such studies would be nearly impossible with mouse models, where the fetus develops inside the mother, making direct observation very problematic.

Zebrafish are also increasingly gaining recognition as a model for neurological diseases. This is largely because their brain shares key neurochemical similarities with the human brain. The key features driving brain diseases, such as protein accumulation and neuronal degeneration, can be replicated in the zebrafish central nervous system. For this reason, zebrafish are widely used in the study of neurodegenerative diseases like Parkinson's, Alzheimer's, Huntington's, and multiple sclerosis. Interestingly, zebrafish are also used to study mental health disorders. Since accurate clinical diagnosis is crucial for treating these conditions, developing an animal model involves understanding the molecular mechanisms of the disease and how they affect behavior.

Zebrafish display complex behaviors similar to those seen in rodents and humans, making them a valuable tool for studying disorders like depression and schizophrenia. Furthermore, a range of behavioral testing tools have been developed for zebrafish, allowing researchers to explore topics like addiction to alcohol and opioids, memory formation, and learning. Additionally, many tools developed for behavioral tests make zebrafish an effective model for studying addiction mechanisms to substances like alcohol and opioids, as well as memory formation and learning.

The striped athlete

Myopathies are a group of diseases where muscle dysfunction leads to weakness. Often these are inherited diseases, caused by genetic mutations. Zebrafish have become a valuable model for studying these disorders, too, largely because their skeletal muscle tissue develops very quickly – it's fully formed within 24 hours of fertilization. Moreover, many of the key molecular processes, such as muscle contraction, energy production, and regeneration, function the same way in zebrafish muscles as they do in human muscles. Since zebrafish larvae start swimming just a day after hatching, researchers can study the effects of mutations on physical activity from the earliest stages of life.

One example of a myopathy is McArdle's disease, caused by a mutation in the gene responsible for breaking down glycogen into glucose, the body's main energy source. As a result, people with McArdle's disease struggle to produce the energy their muscles need, leading to reduced physical endurance and painful muscle cramps after exertion. The disease affects about 1 in 100,000 people, and so far, no effective treatment has been found. To address this, a research team at the University of Wrocław, led by Dr. Marta Migocka-Patrzałek, is developing a zebrafish model for McArdle's disease. Using a fine needle, researchers introduce mutation-causing factors into zebrafish embryos. Early results suggest that this model will not only deepen our understanding of the disease but also help in the search for effective treatments.

Zebrafish are a prime example of a model organism used in scientific research. Their popularity is due to unique characteristics such as their transparent bodies, external development, and short life cycle, making them a valuable alternative to more commonly used organisms like mice or rats. The growing number of scientific publications on zebrafish research is a testament to their increasing importance in the field.

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Fig. 1
Zebrafish embryos encased in semi-transparent egg membranes

Fig. 2
48-hour-old zebrafish larvae

Further reading:

Adhish M., Manjubala I., Effectiveness of zebrafish models in understanding human diseases – A review of models, *Heliyon* 2023, vol. 9 (3), https://doi.org/10.1016/j.heliyon.2023.e14557