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Review

Recent advances in transcriptome research in fishes: A review

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ABSTRACT. Transcriptomic studies have allowed researchers to examine gene activity across the genome and gain insight into many areas of fish biology. This review, following PRISMA guidelines, synthesizes recent studies on fish development, aquaculture, physiology, immunity, nutrition, toxicology, and evolution. It also compares the species studied and the methods used and highlights the biological pathways involved. We reviewed studies that used transcriptomic profiles to trace molecular pathways involved in development, reproduction, and sex differences in farmed fish. Physiological stress studies found that temperature, low oxygen, salinity, pH, and crowding were common stressors. They also showed how these conditions changed gene activity in fish. In immunity and nutrition studies, transcriptomic analyses revealed how fish responded to bacterial challenges and how functional feed additives affected those responses. Toxicology studies linked contaminants to target organs, altered biological pathways, and biomarkers. Finally, transcriptomic data helped researchers distinguish populations, measure evolutionary distances, and identify conserved genes associated with trait differences. Together, these findings show how transcriptomics can help us understand species biology, improve aquaculture, and guide conservation. They also point to gaps that future research should address.

Keywords: transcriptomics; RNA-seq; metabolic pathways; physiology; fish

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INTRODUCTION

Transcriptomics studies all RNA molecules present in a biological sample at a given time and offers insights into cellular functions and gene expression (Page & Lawley 2022). The introduction of next-generation sequencing (NGS), especially RNA sequencing (RNA-seq), has greatly boosted transcriptomic research. This method enables large-scale characterization of gene expression, transcript abundance, and alternative splicing, even in non-model organisms (Deshpande et al. 2023). RNA-seq directly sequences RNA-derived molecules, enabling more comprehensive transcriptome profiling than microarrays, which depend on preconfigured probes for known transcripts (Lee et al. 2024). As a result, RNA-seq is now widely used in transcriptomic research (Ghazanfar 2024).

Sequencing platforms differ in read length and their ability to identify transcripts. For example, Illumina generates short, highly accurate reads and is widely used to quantify gene expression. In contrast, long-read technologies such as PacBio and Oxford Nanopore Technologies generate much longer reads, enabling identification of full-length transcripts and alternative isoforms (Pardo-Palacios et al. 2024, Xu et al. 2024).

As a result, transcriptomics studies in fishes have grown rapidly, producing large amounts of RNA-seq data. The Fish Gene Expression and Transcriptome database (FishGET) collects and curates fish RNA-seq data and provides an annotated reference transcriptome (Guo et al. 2023). Another platform, FishExp, lets users study gene expression and alternative splicing in fish and submit their own data (Tan et al. 2022). Researchers have used transcriptomic profiling to study how fish tolerate salinity stress (Chutia et al. 2022). Transcriptomics can also show how fish respond to environmental changes, such as pH shifts (Fu et al. 2021a). Non-lethal transcriptomic sampling is becoming more common and can provide insights into the physiological status of wild fishes (Jeffries et al. 2021). Overall, transcriptomics studies offer valuable insights into gene expression and molecular responses in fishes, contributing to a better understanding of their biology. These studies also show how fish respond to different pathogens (Sudhagar et al. 2018) by identifying key immune-response genes. For example, in zebrafish testes, glutamine and succinic acid were the most abundant metabolites released, and 27.5% of the genes were related to the immune or reproductive systems (Gelderen et al. 2023).

In flounder, transcriptomic analysis identified genes differentially active in innate immune pathways, including the Phagosome and Nuclear Factor of kappa B (NF-κB) signaling pathways (Wu et al. 2022a). Additionally, transcriptome analysis in Atlantic salmon (*Salmo salar*) identified genes involved in immune responses, including Toll-like receptors and cytokine-cytokine receptor interactions (Andresen et al. 2019). These findings show how transcriptomics can elucidate fish immune responses and identify the genes involved (Ye et al. 2018).

In fish, the transcriptome changes across environmental conditions (Oomen & Hutchings 2017). For example, exposure to elevated temperatures during embryogenesis can alter how tissues adapt later in life and reorganize transcriptomic networks, making gene interactions less organized (Ripley et al. 2023). Additionally, changes in environmental pH can disrupt gene expression in fish, commonly affecting immune-related genes (Fu et al. 2021b). Salinity stress can also alter gene expression in fish, affecting pathways involved in oxygen transport, immune responses, and metabolism (Zhu et al. 2025). Overall, environmental factors can shape the fish transcriptome, driving differential gene expression and potential adaptations to these conditions. Although previous reviews have addressed specific applications of transcriptomics in fish, an integrated synthesis of the species studied, methodological approaches, biological pathways, stress responses, and applications across diverse research areas remains limited. Therefore, this review examines transcriptomic research in fish from 2018 to 2025, focusing on major methodologies, applications, biological responses, emerging trends, and knowledge gaps. The review addresses the following question: what have been the main applications, methodologies, trends, and knowledge gaps in fish transcriptomic research during 2018-2025. The review emphasizes development, aquaculture, physiology, immunology, nutrition, toxicology, and evolutionary genetics to identify common patterns, discrepancies, and potential applications in fish biology, aquaculture, and conservation.

METHODOLOGY: META-ANALYSIS FOLLOWING THE PRISMA GUIDELINES

Selection process

Articles were retained if they (i) constituted original research or systematic reviews applying RNA-seq or transcriptomic approaches to fishes, (ii) were published in peer-reviewed journals, (iii) were published between 2018 and 2025, and (iv) provided extractable methodological and biological information (sequencing

platform, target tissue, species, and main findings). Articles without full-text access, conference abstracts without published data, and non-refereed literature were excluded. Five thematic domains were pre-specified: aquaculture and food safety, physiological stress, immunology, toxicology, and evolutionary genetics. The pre-existing categorical labels in the source review (aquaculture; toxicology and pathogens; physiological stress; nutrition and immune system; genetics) were mapped onto these five themes as follows: aquaculture → aquaculture and food safety; physiological stress → physiological stress; nutrition and immune system and the immunology subset of toxicology and pathogens → immunology; toxicology and pathogens (toxicological subset) → toxicology; genetics → evolutionary genetics.

Information sources and search strategy

A systematic literature search was conducted across databases, including CONRICyT, SciELO, RedALyC, Google Scholar, Scopus, PubMed, Springer, and Web of Science, using standardized search terms related to transcriptomics, RNA-seq, and fish species. The search strategy used Boolean operators to combine keywords such as "transcriptome," "RNA-seq," "gene expression," and "aquatic organisms," along with specific strings for key aquaculture species. The search protocol was designed to ensure comprehensive coverage of the diverse genomic assemblies now available for commercially significant teleosts, capturing data relevant to both laboratory and field-based inquiries. Synthesizing these datasets addresses the critical challenge of reconciling variability across studies, as meta-analyses provide a robust framework for identifying reproducible differential expression patterns and transcription modules that transcend individual experimental designs (Krasnov et al. 2021). To ensure reproducibility and transparency in the synthesis of the selected data, the screening process and the final selection of included studies were documented using a flow diagram in accordance with the PRISMA statement (Nakagawa et al. 2017).

Selection process

Titles and abstracts were screened for thematic relevance. Full texts were then assessed against the eligibility criteria. Data-extraction tables recorded the reference, species, sequencing platform, country, target organ, and main results.

Data extraction and synthesis

For each included study, methodological variables (platform, library preparation, RNA-extraction method, and quantitative reverse transcription polymerase chain reaction (qRT-PCR) validation) and biological outcomes were extracted. Because the included corpus is heterogeneous in platform, tissue, species, and stressor, we synthesized findings narratively by thematic domain, using quantitative descriptors drawn directly from the source review.

Risk-of-bias and methodological considerations

Potential sources of bias included repeated use of TRIzol for RNA extraction and reliance on reference genome quality, particularly for non-model species.

PRISMA flow diagram

flowchart TD

A ["Records identified through database searching

with the terms 'transcriptome and fishes' (2018–2025)"] --> B["Records after duplicate removal screened by title and abstract"]

B --> C ["Full-text articles assessed for eligibility
(full-text access required)"]

C --> D ["Studies included in the meta-analysis
(n = 150)"]

C --> E ["Full-text articles excluded
(non-refereed literature or without full-text access)"]

D-->F ["Thematic synthesis:
Aquaculture & food safety
Physiological stress
Immunology
Toxicology
Evolutionary genetics"]

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Caption. PRISMA-style flow diagram reconstructing the selection process applied in the source review (n = 150 articles, 2018-2025).

A total of 150 articles were included in the meta-analysis procedure (Table 1). The corpus reports the following distribution of affected biological pathways: apoptosis 21.1%, metabolism 18.8%, cellular stress 17.3%, cell signaling 15.8%, and immunity 14.3%.

Methodologically, 89% of the included studies used the Illumina platform in a paired-end configuration, with the Illumina HiSeq 2000 and HiSeq 2500 the most commonly used sequencers. RNA was predominantly extracted with TRIzol®. Library preparation used the Illumina® TruSeq RNA Sample Prep Kit v2 and the NEBNext® Ultra™ RNA Library Prep Kit. qRT-PCR validation was performed in 63% of the studies, with *β-actin*, *ef1α*, *gapdh*, and *18S* most frequently used as reference genes.

Table 1. Study characteristics and quantitative synthesis of the meta-analysis. *The toxicology and pathogens category (35 articles) and the nutrition and immune system category (26 articles) jointly contain immunological studies; partitioning the immunological subset across these two parent categories is not reported in the source review and therefore is not presented here.

Theme	Source-review category	Articles (n)	Percentage (%)
Aquaculture and food safety	Aquaculture	37	24.67
Physiological stress	Physiological stress	29	19.33
Toxicology	Toxicology and pathogens (toxicological subset)	35*	23.33*
Immunology	Nutrition and immune system + immunology subset of toxicology and pathogens	26*	17.33*
Evolutionary genetics	Genetics	23	15.33
Total	-	150	100

Cross-theme methodological observations

- **Aquaculture and food safety** drove the largest category, with strong reliance on RNA-seq from muscle, liver, and gonad to support breeding and nutritional management.
- **Stress studies** were disproportionately molecular, with metabolism, apoptosis, and oxidative stress pathways most represented; immune pathways were altered across stressor types.
- **Immunology studies** increasingly combined host RNA-seq with microbiota profiling, and head kidney/ spleen organs dominated, consistent with the integrative immunomics framework.
- **Toxicology studies** converged on liver transcriptomics, consistently implicating oxidative stress and immune pathways.
- **Evolutionary genetics studies** have opened avenues in non-model species but have also highlighted the absence of conserved transcriptional programs underlying complex behavioral or life-history traits across distant taxa.

Limitations

This synthesis inherits the limitations of the underlying 150-article corpus: (i) Illumina-platform overrepresentation (89%) introduces platform-specific bias; (ii) partitioning the immunology subtopic between nutrition and immune system and toxicology and pathogens categories is not reported in the source review and could not be reconstructed here; (iii) qRT-PCR validation occurred in 63% of the studies, leaving a substantial fraction with expression-level uncertainty; (iv) non-model species frequently lack high-quality reference genomes, limiting pathway resolution despite the feasibility of *de novo* assembly.

This review conducted a systematic literature review of available sources on fish transcriptomes. A semantic search was performed using the terms "transcriptome and fishes" across various national and international digital databases, including CONRICyT, SciELO, RedALyC, Google Scholar, Scopus, PubMed, Springer, and Web of Science. The inclusion criteria covered 150 articles published between 2018 and 2025, including original research and reviews. All documents without full-text access and non-refereed literature were excluded. Subsequently, the articles that met the established criteria were analyzed to systematize them by constructing data extraction tables that integrated relevant information, including reference, species studied, sequencing platform (transcriptomic methodology used), country, target organs, and main results (Fig. 1, and Suppl. Mat.). This review improved comparative research, identified recent transcriptomic trends, and highlighted gaps in current knowledge.

RESULTS

Aquaculture and food security

It was found that 24.67% of the corpus (n = 37) addressed aquaculture and food security; these investigations primarily prioritize optimizing growth metrics and identifying biomarkers for stressors such as environmental temperature fluctuations or overstocking, which are essential for sustainable production practices (Rebl et al. 2020). Furthermore, integrating high-throughput transcriptomics with validated quantitative

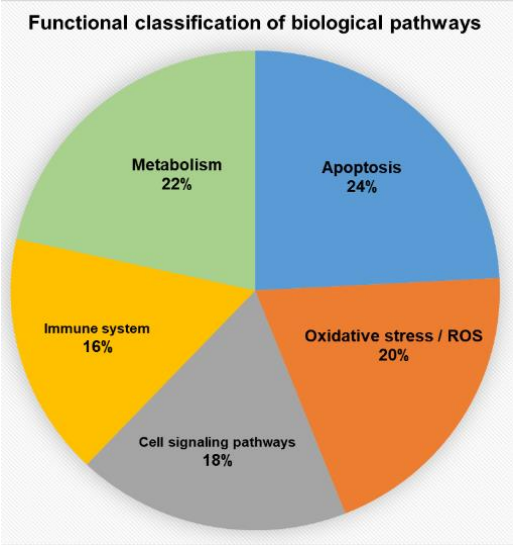


Figure 1. Categories of the biological pathways most affected in transcriptomic studies of fish.

real-time PCR parameters helps refine these welfare indicators, enabling more precise monitoring of fish health in aquaculture environments (Rebl et al. 2020).

In this sense, food insecurity harms many communities worldwide (Yılmaz & Günal 2023). Demand for affordable, high-quality protein is growing (FAO 2023). Aquaculture can help meet this need by supplying nutritious food and supporting economies worldwide (FAO 2022). Despite research efforts, information gaps remain that must be addressed to optimize the aquaculture industry (Xu et al. 2017).

Key aquaculture challenges include improving growth and fillet quality (Zenger et al. 2019). To address these challenges, high-throughput sequencing technologies have been integrated, allowing elucidation of the molecular mechanisms underlying growth and analysis of transcriptomic profiles derived from muscle tissue (Lin et al. 2022). An example of this is found in *Epinephelus fuscoguttatus*, a commercially important species in China, whose muscle transcriptome revealed enriched pathways, including glycolysis/gluconeogenesis, adipocytokine signaling, mitogen-activated protein kinase (MAPK), carbon metabolism, and PI3K-Akt, all of which are involved in growth. Likewise, *ghr*, *tnni2*, *stc2*, and *pdp1* were identified, and their expression was associated with growth advantages in the organisms analyzed (Wang et al. 2023). Fish fillet integrity, structure, and nutritional value also matter (Ali et al. 2018). In *Catla catla*, a muscle transcriptome analysis identified molecular pathways and genes (*myh*14, *acta*1b, and *ghr*2) that regulate growth and could serve as growth biomarkers for breeding programs (Mohindra et al. 2022). Similarly, in the Mexican silverside (*Chirostoma estor*), a stomachless fish, researchers found that feeding frequency changes liver gene activity even without changes in total feed intake. In fish fed 12 times a day, genes linked to metabolism, digestion, growth, cell death, immunity, and oxidative stress were more active, which helps explain their faster growth. In addition, for the first time, liver genes involved in appetite regulation were identified, showing that the transcriptome reflects the molecular mechanisms that control feeding and nutritional physiology (Juárez-Gutiérrez et al. 2024). These findings show how transcriptomics can clarify feeding effects in aquatic species.

Early fish development, by contrast, includes stages of greater susceptibility as organisms undergo numerous changes in body structure and function (Sarropoulou et al. 2014). For this reason, we need to understand what controls early development and reduce the high mortality seen during production (Anderson et al. 2018). Some molecular studies in fish have integrated techniques such as RNA-seq to analyze transcriptomic profiles during the early development of organisms, as done in *Ictalurus punctatus*, from which biological samples were sequenced from days 2, 5, 6, 7, 8, 9, and 10 post-fertilization, and gene expression changes were analyzed (Ma et al. 2020). Different biological processes were identified involving *gdf10*, *foxa2*, *hcea*, *syce3*, *ck1*, *oaz2*, *dars1*, and *ube2v2*, which are associated with head formation, cell cycle, growth, differentiation, and proliferation of enterocytes. Relatedly, these gene-activity profiles can also help predict traits in fish populations (Meier et al. 2014).

One example comes from the Eurasian perch (*Perca fluviatilis*). Researchers built a transcriptome from newly hatched larvae in 16 families. The authors conclude that the transcriptomic profiles provide information on both past events (highlighting the genetic contribution of the broodstock) and offspring development (Debernardis et al. 2025). Likewise, researchers have tracked gene activity during early development in native aquaculture species from southeastern Mexico, such as *Atractosteus tropicus* and *Amphilophus trimaculatus*; both studies identified candidate genes for nutritional markers involved in digestive system morphogenesis and nutritional processes (Hilerio-Ruiz et al. 2021, Martínez-Burguete et al. 2021). It should be noted that the information obtained from this type of research is not only of interest to improve the culture protocols of the species, but also to lay the foundations for the understanding of how their bodies work, which could be applied in the management of natural populations and sustainable fishing activities (Anderson et al. 2018, Liang et al. 2022).

Reproduction is another major theme in fish research -strategies and mechanisms vary widely across species, making it complex. Even so, this work pays off: it informs reproductive management design for the challenges fish face in captivity (Watanabe et al. 2018, Wu et al. 2023). Today, transcriptomes from many species show clear sexual differentiation profiles. Most studies have focused on gonads and the brain (Shen et al. 2020). An example of this was carried out in *Larimichthys crocea* and *Opsariichthys bidens*, in which gonad samples were sequenced. Gene expression profiles identified molecular markers (*dmrt1*, *gsdf*, *sox9*, *amh*, *foxl2*, *dazl*, *dnd*, *nanog*, *piwi*, *vasa*, *zp4*, and *cyp19a*) of sexual differentiation (Xiao et al. 2019, Tang et al. 2022). In *Tor putitora*, the brain tissue of males and females was identified using a transcriptomic approach, which showed a differential expression of genes in both sexes, highlighting the overexpression of *amh*, *foxl3*, *dax1*, *kif20*, *tkt*, *cyp19a1a*, *dmrt2a*, *gdf9*, *sox9b*, *wt-1a*, and *aqp1*, associated with reproduction (Shahi et al. 2023).

Physiological stress

The meta-analysis showed that 19.33% of the corpus (n = 29) examined physiological stress. The synthesis followed PRISMA guidelines to identify key transcriptional responses to environmental stressors, such as temperature, salinity, and hypoxia (Qian et al. 2013, Du et al. 2021). These analyses show how gene expression responds to environmental shifts, underscoring the need for diverse datasets to fully capture aquatic species' adaptive potential across varying ecological conditions. By integrating RNA-seq data, this synthesis shows how these stressors modulate genes involved in immune function and growth (Bernatchez et al. 2017), providing critical insights for managing farmed stocks under future climate change scenarios (Beemelmanns et al. 2021).

Accordingly, several factors can disrupt fish homeostasis. They disrupt homeostasis-the body's internal equilibrium-and trigger the stress response (Schreck & Tort 2016). Today, transcriptomics is a powerful tool. It lets us zoom in at the molecular level and see which genes and pathways shift when fish face stress (Qian et al. 2013). That said, many transcriptomic studies have examined how environmental variables such as temperature, salinity, and dissolved oxygen affect fish (Dettleff et al. 2022, Chen et al. 2023, Lu et al. 2023). Likewise, other factors can stress fish, particularly in farm-raised species, due to inadequate management, such as density, light, and noise (Zhong et al. 2022, Zhou et al. 2023a).

Transcriptomic analyses commonly study the liver, kidney, and gills in thermal stress research across species such as *Megalobrama amblycephala*, *Takifugu rubripes*, *Glyptosternum maculatum*, and *Labrus bergylta* (Li et al. 2018, Liu et al. 2022, He et al. 2023, Palma et al. 2025). By contrast, high-salinity studies mainly used liver tissue, as in transcriptome analyses of *Seriola dumerili* (Ru et al. 2023) and *Scatophagus argus* (Chen et al. 2023). In contrast, transcriptomic techniques identified molecular markers in gill tissue of *Acanthogobius ommatur*s (Sun et al. 2020). In transcriptomic studies evaluating the effects of hypoxia, researchers frequently examine the brain (Shang et al. 2022).

In general, transcriptomic studies that investigated the effects of the different types of stress on fish show that the most affected pathways are metabolic pathways (particularly those related to energy metabolism, protein and amino acid metabolism, lipids and fatty acids, carbohydrates), pathways associated with the immune system, pathways associated with apoptosis, and those involved in oxidative damage (Li et al. 2023a). Researchers also identified several stress-related genes in Nile tilapia (*Oreochromis niloticus*) using Illumina MiSeq sequencing. That study showed that chronic stress harmed the fish: the pathways related to energy metabolism, oxidative damage, and the immune system were the most affected. The study also identified these molecular markers: potassium voltage-gated channel subfamily A regulatory β subunit 1b

(overexpressed); myosin 7; and acyl-coenzyme A thioesterase 4 (underexpressed) (Mwaura et al. 2023).

In another study, *Genypterus chilensis*, a native Chilean fish, showed hepatic oxidative damage and transcriptional modulation in response to thermal stress. Researchers sequenced the liver transcriptome and found 1,239 underexpressed and 1,339 overexpressed genes (negative and positive regulation, respectively), involved in antioxidant responses, including heat shock proteins (hsp60 and hsp70), DNA damage-inducible transcript 4 (ddit4), glutathione peroxidase 7 (gpx7), and leptin (LEP) (Dettleff et al. 2022).

Currently, efforts to understand how fish respond to acute or chronic stress aim to develop biotechnological strategies and tools that mitigate these adverse effects, either in natural populations or on farms, to offset energy costs associated with environmental changes and improve aquaculture efficiency (Souza et al. 2019).

Immunology

In the non-partitioned combined contribution, immunological studies were distributed between the nutrition and immune system category (17.33%) and the immunology subcomponent of toxicology and pathogens (23.33%). Together, these findings suggest that pathogen-induced challenges often trigger signaling cascades shared with husbandry-induced stress, particularly involving Toll-like receptors and the major histocompatibility complex. This meta-analysis highlights the need for large-scale sequencing to delineate shared responses across species, ultimately helping identify prognostic markers for various fish diseases (Türker 2012).

Researchers study fish immune systems (innate and adaptive) because they protect fish health (Uribe et al. 2011). In this context, the intestine is a key defense site against pathogens and responds to physiological challenges from antinutritional factors in alternative plant-based diets (Martin et al. 2016). Molecular approaches and transcriptomic tools help us understand fish immunity (Sudhagar et al. 2018).

Various studies examine tissue structure and function, such as the mucosa and intestinal epithelium, and identify the molecular mechanisms underlying nutrient absorption and defense (Schug et al. 2019). Researchers found a similar pattern in captive *Seriola lalandi*. They used 16S rDNA sequencing to track microbes on the intestinal mucosa and RNA-seq to measure the host response. Enteritis changed the fish microbiota and altered host gene expression and metabolic pathways (Legrand et al. 2020).

Likewise, in the hybrid grouper (*Epinephelus fuscoguttatus* × *E. lanceolatus*), replacing animal protein in the diet with soybean meal led to enteritis in the fish studied. Intestinal transcriptomic analysis revealed changes in genes associated with cell signaling, infectious diseases, and immune responses (Zhang et al. 2021).

Other studies have examined factors such as salinity to understand how fish adapt. Large-scale sequencing has helped researchers assess how brackish and marine water affect the gut microbiota and immune-response genes. Researchers studied *Acanthopagrus latus*, an aquaculture species of interest in Asia, to assess how salinity changes affect its intestinal microbes and genes linked to metabolism and immunity. The results show that brackish water favors *Cetobacterium*. At the same time, seawater promotes *Vibrio* abundance, suggesting potential benefits of brackish-water aquaculture (Cheng et al. 2025). Additionally, the intestinal transcriptomic profile revealed activation of metabolic genes such as *cpa1*, *adh1*, *cyp46a1*, and *slc26a3*, as well as immune-related genes including *ils* and *vdac2* (Peng et al. 2025).

Similarly, researchers have studied the head kidney, a lymphoid organ, in teleost fish to understand its role in immunity (Sun et al. 2022). In fish such as *Eleginops maclovinus* and *Gadus morhua*, transcriptomic profiles of this organ revealed genes linked to both innate and adaptive immunity (Martínez et al. 2018, Solbakken et al. 2019). Transcriptomic resources have also helped researchers develop biotechnological tools for immunology. These tools clarify the molecular pathways that shape immune responses and defense in different fish species (Lazado et al. 2023, Zhou et al. 2024a). For example, researchers study how diet affects fish health by adding functional additives and microorganisms to feed (pre-, pro-, and synbiotics). The aquaculture industry uses these supplements as immunostimulants to strengthen immune responses and support the health of farmed organisms (Cai et al. 2022, Licitra et al. 2023).

Toxicology

Toxicological studies constituted 23.33% of the corpus (n = 35). The liver was the most frequently targeted organ, reflecting its roles in detoxification, energy metabolism, and innate immunity. In this sense, aquaculture provides food and livelihoods for millions of people worldwide. But water quality remains a major challenge in production systems. Primary pollutants include those from mining and the oil industry, insecticides, herbicides, disinfectants, personal care products, antiseptics, and nitrogenous compounds

generated by organisms' metabolism (Caban & Stepnowski 2021). Many of these compounds cause toxicity, environmental degradation, and increased disease susceptibility in aquatic organisms (Yusoff et al. 2024).

New bioinformatics tools and large-scale sequencing now let researchers use transcriptomics to study physiology, detect molecular changes, and clarify how harmful agents act (Kumar et al. 2024, Zhou et al. 2024b). Studies in fish show how toxic contaminants affect aquatic ecosystems, with the liver standing out because it helps detoxify the body, process nutrients, and support immune responses (Yadetie et al. 2021, Zhang et al. 2023). The liver also supports innate immunity by expressing genes involved in pathogen recognition, the complement system, and phagocytosis (Qi et al. 2021). Transcriptomic and histological evidence also shows that the liver contains immune cells and activates immune pathways, making it important for understanding fish physiology, health, and ability to adapt to harsh environmental conditions (Hussein et al. 2023).

Recent RNA-seq studies report liver-related transcriptional changes and changes in pathways linked to immunity, metabolism, oxidative stress, and apoptosis in species such as *Lateolabrax maculatus*, *Micropterus salmoides*, and *Gambusia affinis*, exposed to zinc (Zn), lead (Pb), and triclosan (TCS) as contaminants, respectively (Qian et al. 2019, Zhou et al. 2019, Song et al. 2021). Similarly, a study in *Pseudobagrus ussuriensis* evaluated the effects of acute ammonia exposure using transcriptomic, histological, and biochemical tools. The results showed that ammonia causes severe liver damage, oxidative stress, and genomic changes in this species (Shi et al. 2025).

Transcriptome analysis also helps us understand complex physiological pathways, including immune responses. In recent years, researchers have used RNA-seq to study bacterial, viral, parasitic, and oomycete pathogens in fish (Sudhagar et al. 2018, Maekawa et al. 2019, Mauer et al. 2020, Gao et al. 2022, Chapagain et al. 2023). Researchers analyze the spleen and head kidney because these lymphoid organs play key roles in the immune system (Natnan et al. 2021). An example of this was reported in rainbow trout (*Oncorhynchus mykiss*), where researchers examined spleen transcriptomes after infection with infectious hematopoietic necrosis virus (IHNV) and identified six key genes (*tlr2*, *tlr5m*, *trim25*, *ripk2*, *a20*, and *socs2*) linked to immune responses in the sequencing data (Pan et al. 2023).

Likewise, in rohu (*Labeo rohita*), RNA-seq characterized the head kidney transcriptome after exposure to *Aphanomyces invadans*, revealing differential expression of several immune-related genes. Highly expressed genes were primarily associated with innate and adaptive immune processes, stimulus responses, key signaling pathways, and programmed cell death. These included genes involved in Toll-like receptor and chemokine signaling pathways (Pradhan et al. 2020).

In this way, these gene expression patterns not only identify immune genes but also show the signaling pathways and cellular processes activated in response to infection (Bai et al. 2022, Teng et al. 2022, Huyben et al. 2023, Yang et al. 2023). In this context, transcriptomics has become an essential approach to understanding pathogenesis and host-pathogen interactions in fish, providing a molecular basis for designing effective disease prevention and management strategies in aquaculture (Wu et al. 2019, Angsujinda et al. 2020, Casuso et al. 2022, Huang et al. 2022).

Evolutionary genetics

Evolutionary genetics accounted for 15.33% (n = 23). These studies focus on the diversification of immune-related gene families and species' adaptive molecular evolution in response to distinct environmental pressures (Malmstrøm et al. 2016). To systematize the growing volume of data generated in this field, a meta-analysis was conducted in accordance with the PRISMA guidelines, enabling a robust synthesis of the molecular mechanisms conserved across various teleost species (Carvalho et al. 2012, Ali et al. 2014, Yáñez et al. 2014, Fraslin et al. 2020, Deng et al. 2022).

Transcriptomic resources now generate large datasets that help us understand evolutionary history and interspecific distances (Seixas et al. 2023). These data come from RNA-seq, which allows us to compare gene expression profiles. These profiles can indicate adaptive responses, resilience, and the physiological processes that help organisms cope with environmental conditions (Connon et al. 2018). In this context, bioinformatic analyses can reveal molecular mechanisms in species that previously lacked functional genomics resources; an example of this was the research carried out on *Platypharodon extremus*, an endemic species from the Tibetan Plateau that faces extinction; sequencing it allowed researchers to study bone morphogenetic proteins (BMPs), which help explain its development and the genes involved (Wu et al. 2022b).

In *Carassius auratus*, transcriptomic sequencing showed that artificial selection changed goldfish gene

Table 2. Species used as classical biological models through transcriptomic studies.

Species	Environment	Countries	References	Study
Danio rerio	Freshwater	Sweden, USA Singapore, UK China, Italy	Porseryd et al. (2018) Boswell et al. (2019) Li et al. (2022) Ripley et al. (2023) Liu et al. (2023) Licitra et al. (2023)	Genetic and developmental model
Takifugu rubripes	Marine, freshwater, and brackish	China	Liu et al. (2022) Shang et al. (2022) Lu et al. 2023)	Genomic model
Oryzias latipes	Freshwater and brackish	China	Wang et al. (2024)	Genetic and developmental model
Carassius auratus	Freshwater and brackish	China	Pang et al. (2018) Du et al. (2023)	Model in neurophysiology, vision, and toxicology

expression and, in turn, their genome (Du et al. 2023). Likewise, researchers studied distantly related species, including *Danio rerio*, *S. salar*, and *Dicentrarchus labrax*, to test whether a conserved molecular pathway links physiology and behavior through the brain transcriptome. The study found no shared gene-expression signature or pathway for proactive behavior across these species' genetic networks and molecular functions (Rey et al. 2021).

In model species such as *D. rerio*, researchers have developed extensive genetic resources, including genome and transcriptome sequencing, that help them study the functions of specific genes. These resources help identify transcription factors and key molecular pathways involved in cell growth and regeneration (Aanes et al. 2011, Lee et al. 2020). They provide knowledge that supports biomedical research in humans (Sandler et al. 2025). Together, these findings show that transcriptomics applies across fields. In fish evolutionary genetics, researchers have studied topics ranging from functional genomics and evolution to conservation biology (Connon et al. 2018). These studies have generated biologically and physiologically valuable information for genetic and population management in cultivation and natural ecosystems, as well as for biotechnological applications in areas such as medicine and pharmacology.

In this review, the species were classified as classical, applied, or emerging models based on their biology, genetics, development, research output, ecological relevance, genomic and transcriptomic resources, and uses in aquaculture, biotechnology, and medicine (Srivastava et al. 2025). Among the models used, *S. salar*, *O. mykiss*, and *O. niloticus* were the most studied. Among the emerging models, *Colossoma macropomum*, *Cyprinus carpio*, *Acanthopagrus schlegelii*, *Siniperca chuatsi*, and *Seriola dumerili* stand out. In contrast, *D. rerio* is the most representative and classical biological model, given the large number of studies worldwide (Table 2).

Among the sequencing platforms used in the reviewed fish transcriptomic studies, the Illumina HiSeq 2000 was the most widely used, followed by the Illumina HiSeq 2500. Other frequently used platforms included the Illumina NovaSeq 6000, the Illumina HiSeq 4000, and the Illumina NextSeq 500, which were among the five most common platforms reviewed (Table 3).

Applied aquaculture was the most common topic among the fish transcriptomic studies reviewed, accounting for 46.77% of the articles. It was followed by toxicology and pathogens (23.33%), physiological stress (19.33%), nutrition and the immune system (17.33%), and genetics (15.33%). These results reflect the diversity of approaches in fish transcriptomic studies and highlight the predominant interest in practical applications in aquaculture and organism health (Table 4).

Looking at biological pathways, apoptosis was the most common category, accounting for 21.1% of cases. It was followed by metabolism (18.8%), cellular stress (17.3%), cell signaling (15.8%), and immunity (14.3%). These findings show that apoptosis and metabolism were the most common pathways, but they also reveal important changes in processes linked to the immune system's stress response. This pattern reflects the range of molecular mechanisms involved in fish physiology and adaptation (Fig. 1). Overall, transcriptomic studies

Table 3. Sequencing platforms most used in transcriptomic studies of fish, indicating the topic, the organ analyzed, and the reference.

Platforms	Topics	Samples analyzed	References
Illumina HiSeq 2000	Aquaculture	Larvae	Martínez-Burguete et al. (2021)
Illumina HiSeq 2500	Toxicology and pathogens	Branchia	Yang et al. (2022)
Illumina NovaSeq 6000	Nutrition and immune system	Intestine and spleen	Toxqui-Rodríguez et al. (2025)
Illumina HiSeq 4000	Physiological stress	Liver	Li et al. (2023)
Illumina NextSeq 500	Genetics	Embryos	Sandler et al. (2025)

Table 4. Number and percentage of transcriptomic publications in fish by research topic.

Topic	Number of articles	Percentage (%)
Genetics	23	15.33
Nutrition and the immune system	26	17.33
Physiological stress	29	19.33
Toxicology and pathogens	35	23.33
Aquaculture	37	24.67
Total	150	100

in fish tend to identify pathways primarily related to cellular regulation and homeostasis, as well as organismal physiological responses (Liu et al. 2025).

Final remarks

Of the 150 articles reviewed, 89% used the Illumina platform with paired-end sequencing for large-scale RNA-seq. The most common models were Illumina HiSeq 2000 and Illumina HiSeq 2500. Both support paired-end sequencing, with read lengths of up to 2×100 bp and 2×125 bp, respectively (Minoche et al. 2011, Schirmer et al. 2016). For RNA extraction from the tissues studied, 89% of the studies used TRIzol® reagent from different brands, including Invitrogen, Ambion, and Sigma-Aldrich. Some articles report checking RNA purity and integrity by running it on an agarose gel or measuring its 260/280 absorbance ratio with spectrophotometry (Caballero-Solares et al. 2018, Tsakogiannis et al. 2018).

On the other hand, for the preparation and/or construction of libraries to be sequenced, the most commonly used kits were the Illumina® TruSeq RNA Sample Prep Kit v2 (Illumina®, San Diego, CA, USA) and the NEBNext® UltraTM RNA Library Prep Kit (Illumina, USA), following the protocols specified by their respective manufacturers (Lazado et al. 2023, Lu et al. 2023). Another debated methodological issue is validating transcriptome gene expression with qRT-PCR. In this review, 63% of the studies report this validation, and some authors even share the genes and primers they used. These include *β-actin*, *ef1α*, *gapdh*, and *18S*, which are considered reference genes, also called housekeeping genes (Li et al. 2018, 2023b, Yan et al. 2022, Liu et al. 2023, Culbert et al. 2025).

Currently, fish studies increasingly integrate methods that study genes, RNA, proteins, and metabolites together, providing a stronger basis for comparison (Infante et al. 2008). One example is a study of *Hemibagrus wyckioides*, a catfish of aquaculture and commercial importance in China. The researchers assembled a chromosome-level genome and sequenced barbel RNA to study these sensory structures' functions. The authors highlight that the study lays the molecular foundations for barbel regeneration and provides new insights into the evolution of feeding habits in this group of fish (Zhou et al. 2023b).

Likewise, researchers used several omics methods in *Epinephelus lanceolatus* to study SGIV iridovirus-induced disease. They examined liver tissue microscopically and measured its RNA, proteins, and metabolites. They found that infection damaged the tissue and changed immune and metabolic pathways. These findings can guide prevention and control of iridovirus disease (Liu et al. 2024).

Today, multi-omics studies use new biotechnological tools to explore areas such as health and well-being, genetic improvement, nutrition, immunity, and stress (Natnan et al. 2021). Together, these approaches help us understand biological systems as a whole (Choudhary et al. 2024). In addition, continued development of bioinformatics tools for each omics approach can help us analyze and integrate these data into a broader view of molecular-level biological and functional networks (Sundaray et al. 2022).

Credit author contribution

A. Hilerio Ruiz & Y. Jiménez León: conceptualization, methodology, formal analysis; C.A. Álvarez González, F.J. Toledo Solís, R. Martínez García, G.G. Asencio Alcudia & D. Tovar-Ramírez: conceptualization, methodology, formal analysis, writing-original draft, review and editing; L.D. Jiménez-Martínez & S. del C. de la Rosa-García: writing-original draft, review and editing. All authors have read and accepted the published version of the manuscript.

Conflict of interest

The authors declare no conflict of interest.

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