

REVIEW ARTICLE OPEN ACCESS

Genomic Insights Into the Use of Green Tea (*Camellia sinensis*) as a Functional Feed Additive in Sustainable Aquaculture

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ABSTRACT

Aquaculture is the fastest-growing sector of animal food production, yet it faces increasing challenges in maintaining fish health and sustainability due to the widespread use of antibiotics and synthetic feed additives. Green tea (*Camellia sinensis*), a natural source of polyphenols and bioactive compounds, has gained attention as a functional feed additive capable of enhancing growth performance, immune function and disease resistance in aquaculture species. This review critically examines genomic and omics-based evidence supporting the efficacy of *C. sinensis* in fish and shrimp nutrition. Findings from quantitative PCR (qPCR) and transcriptomic studies reveal that green tea supplementation significantly modulates the expression of key immune and antioxidant genes, including *IL-1 β* , *TNF- α* , *SOD* and *CAT*, thereby strengthening innate immune responses and oxidative stress resistance. In addition, green tea influences critical stress and pathogen-response pathways through the upregulation of *HSP70*, toll-like receptors (TLRs) and activation of the NF- κ B signalling cascade. These molecular responses collectively contribute to enhanced physiological resilience and reduced reliance on antibiotics. Comparative data indicate consistent improvements in growth metrics, gene expression profiles and immune biomarkers across multiple aquaculture species, emphasizing dose specificity and conserved pathways across species. The integration of green tea into aquafeeds presents a sustainable, eco-friendly strategy aligned with modern functional feed innovations. Future research should aim to optimize dosage levels, evaluate species-specific responses and employ integrative multi-omics approaches to further elucidate long-term effects on aquatic health and environmental sustainability.

1 | Introduction

The aquaculture sector is a significant source of animal protein and serves as an effective means for supplying the expanding world population with the required protein. In 2022, global

aquaculture production reached a record 130.9 million tonnes, comprising 94.4 million tonnes of aquatic animals, accounting for 51% of total aquatic animal production, and 36.5 million tonnes of algae (D'Abramo 2025; Dongyu 2024). Nevertheless, the intensification of aquaculture systems through the growth

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in worldwide trade has raised serious concerns associated with the sustainable development of aquaculture particularly the risks and transmission of infectious diseases in animals (Reverter et al. 2020; Hridoy, Munny, et al. 2025). Non-nutritive additives are being added to the aquafeeds at optimum levels to achieve better nutrient utilization and improve physiological performances (growth, immunity and reproduction) (Mohanta 2024; Hridoy et al. 2024; Das et al. 2024). Traditional additives in animal feed (pellet binders, antioxidants and preservatives) are essential for the stability and quality of feed (Encarnação 2016).

Antimicrobial resistance (AMR) poses a significant threat to the aquaculture industry, particularly in developing countries with warm climates and limited regulatory enforcement (Henriksson et al. 2018). The overuse and misuse of antibiotics in aquaculture systems have led to the emergence of antibiotic-resistant bacteria, which are now prevalent in many foodborne pathogens (Reverter et al. 2020). This issue is particularly alarming as resistant bacteria can transfer to humans through the food chain, exacerbating public health concerns (Suyamud et al. 2024). The growing prevalence of AMR in aquatic food products not only compromises human health but also diminishes the efficacy of antibiotics in treating infections, leading to higher healthcare costs and complicating global efforts to control infectious diseases. Thus, herbal feed supplements are of interest as natural and eco-friendly alternatives to synthetic drugs. These plant herbal additives are made up of natural substances that abound in bioactive compounds (flavonoids and alkaloids), which enhance immunity and growth in aquatic animals with no or less residual and side effect effects (Barad et al. 2024; Kuebutornye et al. 2024; Zhu 2020).

Camellia sinensis is a potential functional feed ingredient for aquaculture, containing bioactive compounds, particularly catechins, that possess anticancer, antioxidant, anti-inflammatory, anti-diabetic, anti-obesity, cardiovascular protection and immunological regulation properties (Luo et al. 2024; Al-Ngada et al. 2017; Zhou et al. 2012). These compounds exert multi-target effects, enhancing immune function and regulating gut microbiota, which plays a critical role in digestion, nutrient absorption and disease resistance. Furthermore, the structure–function relationship of these polyphenols allows them to interact with specific receptors, such as TLR2, thereby influencing immune signalling pathways and promoting cellular defence mechanisms. Tea polyphenols (TPs) are naturally occurring chemicals found mostly in green tea and other type of plants, such as olives. TPs primarily consist of catechins, flavanols, proanthocyanidins and theaflavins that possess diverse biological activities (Daniel et al. 2024).

The significance of *C. sinensis* in aquaculture is demonstrated by its impact on critical molecular pathways related to aquatic animal health. Numerous studies have highlighted that green tea bioactives can regulate the expression of critical immune and antioxidant genes such as interleukin-1 β (IL-1 β), tumour necrosis factor- α (TNF- α), superoxide dismutase (SOD) and catalase (CAT) (Z. Zhao et al. 2023; Mohabbulla et al. 2016). These regulatory effects result in higher immunological responses, lowered oxidative stress and increased resilience to environmental and pathogenic stressors (Ahammed et al. 2023; Prasanth et al. 2019). Moreover, green tea influences the expression of genes involved

in stress and disease response pathways, including heat shock protein 70 (HSP70), toll-like receptors (TLRs) and nuclear factor-kappa B (NF- κ B). As a consequence, it boosts the cellular defence mechanisms of organism (Mokra et al. 2022; C. Y. Chen et al. 2018).

Moreover, green tea supplementation in aquafeeds facilitates sustainable aquaculture by reducing environmental impact, improving fish welfare and decreasing reliance on antibiotics and synthetics (H. Yu et al. 2024; Hernández-Contreras et al. 2023). It is a potential functional ingredient as consumer interest in eco-friendly, traceable products grows. Advances in omics technologies including quantitative polymerase chain reaction (qPCR) and transcriptome analysis have confirmed that it modifies immunological, antioxidant and metabolic pathways at molecular levels (Li et al. 2022; Si et al. 2021). These findings offer empirical and mechanistic evidence in support of the beneficial impacts on fish health and performance, and reinforces the value of green tea as part of sustainable aquafeed formulations.

This review was intended for comprehensively examine the functional potentials of *C. sinensis* as a natural feed additive in aquaculture. It particularly emphasized the contribution of bioactive compounds derived from green tea in influencing immune, antioxidant and stress systems in aquatic organisms. The review highlighted the health-promoting effects of green tea, its role in boosting resistance against pathogens and its contribution to the sustainability of aquaculture, incorporating recent advances from molecular and omics studies. Finally, this review addressed the broader consequences of adopting eco-friendly feed policies in response to global concerns about environmental effect and antibiotic use.

2 | Methodology

2.1 | Scope and Objectives of the Review

This review aims to critically synthesize current genomic and omics-based evidence on the use of *C. sinensis* as a functional feed additive in aquaculture. The scope of the review encompasses finfish and crustacean species commonly used in aquaculture, with particular emphasis on molecular, transcriptomic and functional genomic responses associated with immune modulation, antioxidant defence, stress tolerance and disease resistance. The primary objectives are to:

- i. summarize gene-level and pathway-level responses induced by green tea supplementation
- ii. identify conserved molecular mechanisms across species
- iii. evaluate dose-dependent and species-specific effects
- iv. assess the potential of green tea to reduce antibiotic dependency in aquaculture systems.

2.2 | Literature Search Strategy

A comprehensive literature search was conducted using major scientific databases, including Web of Science, Scopus and

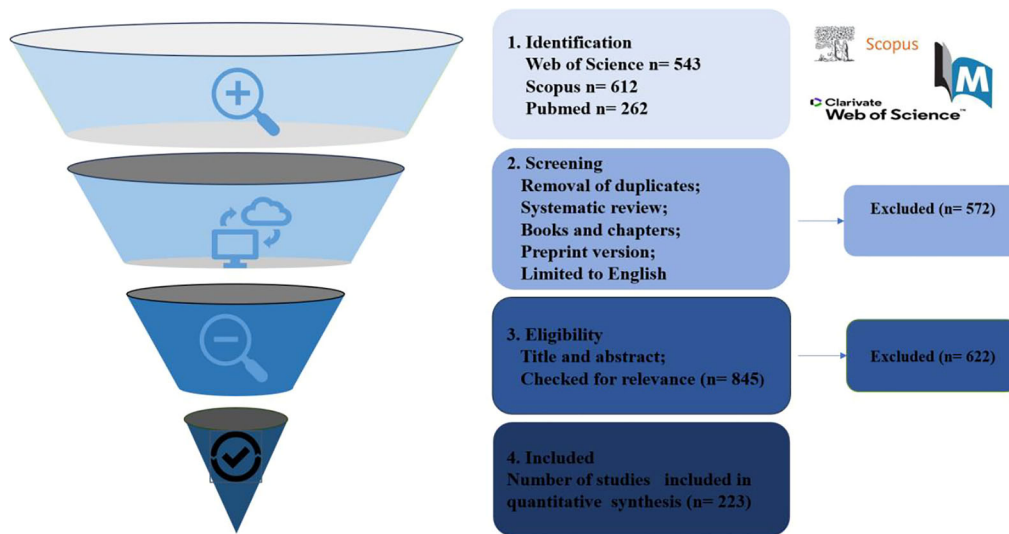


FIGURE 1 | PRISMA structural framework for literature screening process. Non-translated works were excluded.

PubMed. The search covered peer-reviewed articles published up to 2025. Keywords and Boolean combinations included: ‘*Camellia sinensis*’ OR ‘green tea’ AND ‘aquaculture’ OR ‘fish’ OR ‘shrimp’ AND ‘gene expression’ OR ‘transcriptomics’ OR ‘qPCR’ OR ‘omics’ AND ‘immunity’ OR ‘antioxidant’ OR ‘stress response’.

Additional relevant studies were identified through backward and forward citation tracking of key review and experimental papers. Only articles published in English were considered.

2.3 | Inclusion and Exclusion Criteria

The literature screening and selection process followed the PRISMA structural framework, and studies published in languages other than English were excluded from the analysis (Figure 1) (Bordin et al. 2025; Al Mamun Hridoy et al. 2025). Studies were included in this review if they investigated the dietary supplementation of *C. sinensis* or its extracts in fish or shrimp and reported molecular, genomic, transcriptomic, or other omics-based outcomes, such as quantitative PCR or RNA sequencing analyses. Eligible studies were required to assess at least one biological endpoint related to immune function, antioxidant defence, stress response or growth performance and to be original research articles published in peer-reviewed journals.

Studies were excluded if they focused exclusively on terrestrial animals or human nutrition, lacked molecular or gene-level evidence or were limited to conference abstracts, editorials or other non-peer-reviewed sources. In addition, studies examining green tea solely as a waterborne treatment rather than as a dietary feed additive were not considered for inclusion.

2.4 | Data Extraction and Thematic Analysis

From each eligible study, data were systematically extracted on species, life stage, green tea source and dosage, experimental duration, molecular techniques employed, target genes or path-

ways and reported biological outcomes. Extracted data were qualitatively synthesized using a thematic analysis approach.

Gene expression outcomes were categorized into major functional themes, including innate immunity, antioxidant defence, stress response and pathogen recognition signalling. Comparative analysis was performed to identify conserved molecular pathways (e.g., NF- κ B, TLR signalling) and consistent gene expression trends across species and experimental conditions.

2.5 | Quality Assurance and Search Limitations

To ensure data reliability, only studies published in indexed, peer-reviewed journals were included. Methodological rigor was assessed based on experimental design clarity, use of appropriate controls, replication and transparency of molecular techniques.

Nevertheless, certain limitations exist. Variability in experimental designs, dosages, extract compositions and molecular endpoints limited direct quantitative comparisons. In addition, the scarcity of long-term and integrative multi-omics studies constrained the comprehensive evaluation of chronic effects and ecosystem-level implications. Despite these limitations, the available evidence provides robust molecular insights into the functional role of *C. sinensis* in aquaculture nutrition.

3 | Mechanisms of Action

Green TPs, particularly epigallocatechin gallate (EGCG), are gaining attention for their immunomodulatory and antioxidant effects in aquaculture. They modulate gene expression and signalling pathways that enhance fish innate immunity and stress resistance in intensive culture conditions (Deng et al. 2023; Wang et al. 2020). EGCG is a key mediator in oxidative stress and immune regulation, primarily by modulating cytokine expression, activating both non-specific and specific immunity and enhancing antioxidant enzymes (X. Yang et al. 2024; Ahmadifar et al. 2021).

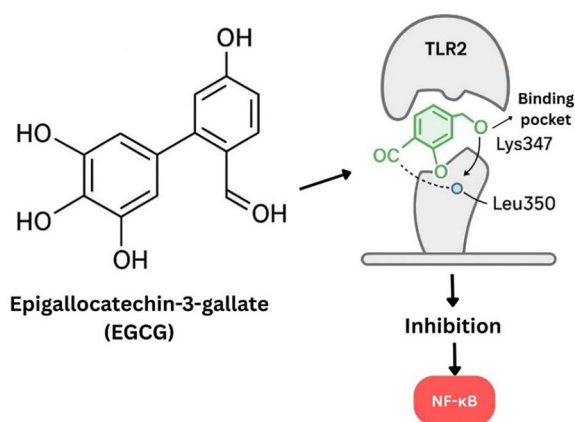


FIGURE 2 | Schematic representation of the EGCG chemical structure and its interaction with TLR2, inhibiting NF- κ B activation in aquaculture species.

3.1 | Immune and Antioxidant Regulation

3.1.1 | Immune Response Genes (*IL-1 β* and *TNF- α*)

In high-density rearing environments, fish are exposed to environmental stressors and pathogens, which can suppress immune responses. Dietary supplementation with green TPs, especially EGCG, has been suggested as an effective method to improve disease resistance and immune function in cultured species (Deng et al. 2023). Dietary EGCG supplementation significantly influenced the expression of immune-related genes, including serum lysozyme (LZM) and IgM levels, enhancing innate immunity in *Monopterus albus* (Deng et al. 2023). It also increased the expression of genes involved in antigen presentation and the antimicrobial immune response, such as hepcidin and MHC-2 α , as well as upregulated IL-8, indicating improved leukocyte chemotaxis. In addition to upregulation of immune-protective markers, EGCG exhibited anti-inflammatory activities by reducing the expression of IL-6 and NF- κ B, key players in pro-inflammatory signal transduction pathways (Ren et al. 2024). In largemouth bass (*Micropterus salmoides*), dietary green TPs had similar downregulatory effects on hepatic IL-1 β production as in rainbow trout (*Oncorhynchus mykiss*), in which EGCG largely decreased systemic immune responses (Nootash et al. 2013). As different fish species require different types of treatment for that reason their expression also changes (Table 1).

In *Cyprinus carpio*, TP promoted both humoral and mucosal immunity by increasing the expression levels of C3, C4 and the immunoglobulins (IgM, IgD, IgT) in response to lipopolysaccharide (LPS) challenge (Espinosa et al. 2020). Furthermore, polyphenols derived from green tea decreased the mRNA expression of TNF- α , IL-6 and IL-1 β in intestinal. Meanwhile, molecular docking results showed higher binding activity against TLR2 for TPs and EGCG as compared to other stimulants, which further confirmed their immunoregulatory potentials. This interaction is expected to inhibit the triggering of TLR2/NF- κ B signalling pathway (M. Qian et al. 2025). EGCG binds within the TLR2 binding pocket, acts on key interacting residues with Lys, Ser and Leu by as shown in Figure 2 which shut down downstream signalling and NF- κ B activation. Such immunostimulatory impacts have

also been reported for crustaceans. EGCG induced the expression of several antimicrobial peptides through the immune deficiency (IMD) pathway in *Marsupenaeus japonicus* (Wang, Li, et al. 2021).

This indicates that EGCG is capable of modulating evolutionarily conserved immune pathways in aquatic organisms. Furthermore, a dose-dependent response was observed in *M. albus*, and the suitable dietary EGCG level feeding diet to enhance IgM production obtained by regression analysis using six graded dietary supplementation levels (0, 25, 50, 100, 200 and 400 mg/kg) which were strategically selected according to their preliminary physiological and toxicological data for *M. albus*. The ability of using a gradient dosing approach in the present study allowed to measure throughout the exposures, at multiple response levels both on immune and antioxidant markers based on which stronger indication towards identifying optimal dose of supplementation. The linear relationships observed between immunological markers with dietary EGCG concentration, as well as the regression-derived optimal dose of approximately 110 mg/kg emphasize the need for species specific gradient dose designs to optimize functional feed additive efficacy while minimizing any adverse effects. (Deng et al. 2023). Green TPs can control immune-related genes such IL-1 β and TNF- α , reducing inflammation and maintaining the immunological condition of aquaculture animals. Therefore, green TPs particularly EGCG have broad profile of immunomodulatory effects through increasing immune markers and reducing pro-inflammatory cytokines. The regulation of these IL-1 β , TNF- α and genes-related indicate a genomic foundation for applying TPs to be a sustainable substitute for antibiotics in aquaculture feed strategies.

3.1.2 | Antioxidant Enzyme Genes (SOD and CAT)

Oxidative stress, resulting from overproduction of reactive oxygen species (ROS), is a major threat to fish health in intensive aquaculture. Fish utilize antioxidant enzymes like CAT and SOD to combat oxidative damage (Hoseinifar et al. 2020). Dietary supplementation with green TPs, particularly EGCG, enhances the expression of antioxidant enzyme genes such as SOD and CAT, boosting enzymatic activity. For example, EGCG supplementation in rainbow trout (*O. mykiss*) increased SOD and CAT activities, enhancing antioxidant defence (Deng et al. 2023). Gene expression profiles of IL-1 β , TNF- α , SOD and CAT in fish fed with green tea-based feed is illustrated in Figure 3.

Likewise, koi carp (*C. carpio*) fed with TP diet had enhanced antioxidant status (Y. Zhang et al. 2022). However, non-specific immunity was slightly inhibited at the same time. While EGCG is one of the most widely studied antioxidants, with greatest potential benefits for low to moderate doses, effects of EGCG are arguably dose-dependent and species specific. *M. albus* and Zhao et al. (2023) have suggested that EGCG supplementation at quantities of around 100 mg/kg may promote antioxidant enzyme activity and improve immune functions Likewise, groups receiving 200 mg/kg or above all showed a significant increase in the levels of lipid peroxidation as revealed by malondialdehyde (MDA) concentration and notable decrease in CAT activity and reduced glutathione level—both important antioxidants. This is probably due to its ability to stabilize ROS production and

TABLE 1 | Comparative effects of dietary green tea polyphenols (EGCG) on immune-related gene expression (IL-1 β , TNF- α , IgM, C3, C4, etc.) across different aquaculture species.

Fish species	Treatment type	Target genes/markers	Expression change	Functional outcome	Significance	References
<i>Monopterus albus</i>	Dietary EGCG: 0, 25, 50, 100, 200, 400 mg/kg	Weight gain and SGR	Higher at 0 and 100 mg/kg	Growth performance	Significant	Deng et al. (2023)
		Whole-body lipid content	↓ at 25, 50, 400 mg/kg	Lipid metabolism		
		AST (aspartate aminotransferase)	↓ in all EGCG groups except 100	Liver function marker		
		CAT (catalase)	↓ as EGCG dose increases	Antioxidant enzyme		
		MDA (malondialdehyde)	↑ as EGCG dose increases	Oxidative stress marker		
		mhc-2 α (major histocompatibility complex class II alpha)	↑ with EGCG	Antigen presentation		
		Hepcidin	↑ with EGCG	Antimicrobial peptide		
		il-8 (interleukin-8)	↑ with EGCG	Pro-inflammatory cytokine		
		il-6 (interleukin-6)	↓ with EGCG	Pro-inflammatory cytokine		
		nf- κ b (nuclear factor kappa B)	↓ with EGCG	Transcription factor		
<i>Oreochromis niloticus</i>	Dietary TPs: 0, 100, 200, 300, 400, 500 mg/kg	Final body weight (FBW)	↓ at 500 mg/kg	Growth performance	$p < 0.05$	Daniel et al. (2024)
		Whole-body moisture content	↑ at 100 mg/kg	Nutritional composition		
		Crude protein content	↓ at 100 mg/kg	Nutritional composition		
		Plasma LDL (low-density lipoprotein)	↑ at >200 mg/kg	Lipid metabolism		
		Plasma TG (triglycerides)	↓ at all TP levels vs. control	Lipid metabolism		
		CAT (catalase)	↑ at all except 300 mg/kg	Antioxidant enzyme		

(Continues)

TABLE 1 | (Continued)

Fish species	Treatment type	Target genes/markers	Expression change	Functional outcome	Significance	References
<i>Micropterus salmoides</i>	Dietary FT: 0%, 2%, 4%, 6%	SOD (superoxide dismutase)	↑ at 200, 400, 500 mg/kg	Antioxidant enzyme	$p < 0.05$	L. Jiang et al. (2022)
		T-AOC (total antioxidant capacity)	↑ at 400–500 mg/kg	Antioxidant capacity		
		GSH (glutathione)	↑ at 500 mg/kg	Antioxidant molecule		
		MDA (malondialdehyde)	↑ at 100–200 mg/kg	Oxidative stress marker		
		WGR (weight gain rate)	↑ at ≥ 2% FT (fermented tea residue)	Growth performance		
		SGR (specific growth rate)				
		VH (intestinal villi height)	↑ at ≥ 2% FT	Intestinal morphology		
		SOD (superoxide dismutase)	↑ at ≥ 2% FT	Antioxidant enzyme		
		GSH-Px (glutathione peroxidase)	↑ at ≥ 2% FT	Antioxidant enzyme		
		CAT (catalase)	↑ at ≥ 2% FT	Antioxidant enzyme		
		MDA (malondialdehyde)	↓ at ≥ 2% FT	Oxidative stress marker		
		Mortality after <i>Aeromonas hydrophila</i> challenge	↓ cumulative mortality with FT	Disease resistance		
		IL-10 (interleukin-10)	↑ with FT after infection	Anti-inflammatory cytokine		(Continues)
		TGF-α (transforming growth factor alpha)	↑ with FT after infection	Anti-inflammatory cytokine		
		IL-1, IL-15, IL-8, TNF-α	↓ with FT after infection	Pro-inflammatory cytokines		

TABLE 1 | (Continued)

Fish species	Treatment type	Target genes/markers	Expression change	Functional outcome	Significance	References
<i>Cyprinus carpio</i>	TP supplementation ± LPS challenge	Lysozyme (LZ)	↑ with TP	Innate immune enzyme	Significant	M. Qian et al. (2025)
		Alkaline phosphatase (ALP)	↑ with TP	Immune enzyme		
		Complement C3, C4	↑ with TP	Immune components		
		IgT, IgD, IgM	↑ with TP	Immunoglobulins (adaptive immunity)		
		TLR2 (toll-like receptor 2)	↓ with TP during LPS challenge	Pattern recognition receptor		
		MyD88 (myeloid differentiation primary response 88)	↓ with TP	Adapter protein in TLR signalling		
		NF-κB p65	↓ with TP	Transcription factor		
		IκBα (inhibitor of NF-κB)	↓ with TP	NF-κB inhibitor		
		TNF-α, IL-6, IL-1β	↓ with TP	Pro-inflammatory cytokines		
		Tight junction proteins (claudins, occludin, ZO-1)	↑ with TP	Intestinal barrier integrity		
<i>Epinephelus fuscoguttatus</i> × <i>E. lanceolatus</i>	TP at 0 (TP1) to 0.16% (TP6) in high-lipid diet (16.7%) for 8 weeks	Phosphorylated MLC (myosin light chain)	↓ with TP (reversed LPS effect)	Cytoskeleton regulation	$p < 0.05$	Pan et al. (2022)
		Growth performance (WGR, SGR)	↑ with TP, highest at 0.04% (TP4)	Growth indicators		
		Protein efficiency ratio (PER)	↑ at TP4	Protein utilization		
		Antioxidant enzymes (SOD, CAT, GSH-Px, T-AOC)	↑ in serum and liver	Oxidative stress defence		
		Oxidative stress markers (ROS, MDA)	↓ with TP	Oxidative damage markers		

(Continues)

TABLE 1 | (Continued)

Fish species	Treatment type	Target genes/markers	Expression change	Functional outcome	Significance	References
		Immune enzymes (ACP, AKP, LYS, IgM)	↑ in liver	Immune response indicators		
		Gene expression (liver): sod, cat, gsh-px, nrf2, keap1, hsp70, hsp90, myd88, tnfr	Upregulated	Antioxidant and immune genes		
		Gene expression (liver): thr22, il8, il1β, il10	Downregulated	Inflammatory markers		
		Gene expression (head kidney): myd88, il1β, tnfr, il6	Upregulated	Immune signalling		
		Gene expression (head kidney): thr22, il10	Downregulated	Immune regulation		
		Survival after <i>Vibrio harveyi</i> challenge	↑ survival in TP4-TP6 groups	Disease resistance		

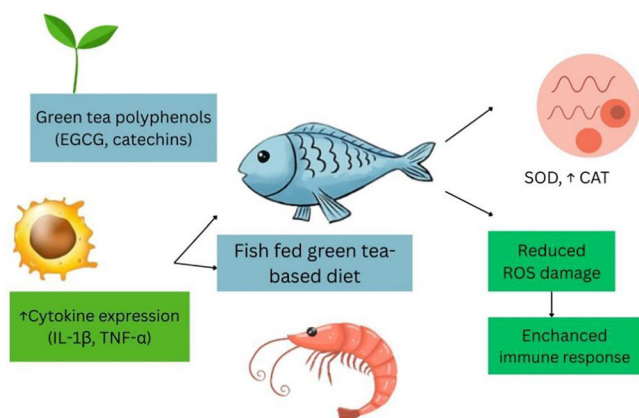


FIGURE 3 | Immune and antioxidant gene expression profiles of IL-1 β , TNF- α , SOD and CAT in fish fed with green tea-based feed. Green tea polyphenols, EGCG, in particular, regulate the expression of cytokines and antioxidant enzymes and thus contribute to immune defence and oxidative stress tolerance significant for aquaculture species.

auto-oxidation at supraphysiological concentrations. The similar pro-oxidative effects in a dose-dependent manner were also noted in *Pseudobagrus fulvidraco* at 100 mg/kg EGCG (Guo et al. 2021). The current study evidenced the importance of establishing a dose threshold of EGCG supplementation for aquaculture species to promote its immunostimulatory actions and prevent oxidative damage. The broken-line regression analysis in *M. albus* further supported this threshold, which was approximately 110 mg/kg dietary EGCG (Deng et al. 2023). Therefore, careful dosing is important for obtaining health benefits and minimizing deleterious pro-oxidant effects.

The Keap1/nuclear factor erythroid 2-related factor 2 (Nrf2) signalling pathway is a major cellular defence system against oxidative stress. However, species and experimental paradigm-specific opposing responses seen as upregulation or downregulation of Keap1 and Nrf2 transcripts have been reported as well across different animal models (even within the same species). This apparent discrepancy is largely due to the ability of TPs to regulate Nrf2 expression through Keap1-independent pathways, thereby confounding traditional activation pathway paradigms (Yang et al. 2025).

For example, hepatic keap1 and nrf2 mRNA levels were increased concurrently by dietary supplementation with 0.02% TPs in largemouth bass (*M. salmoides*). This differs from the conventional paradigm in which that same process of Keap1 suppression by TPs would lead to stabilization of Nrf2 and subsequent induction of antioxidants. These findings suggest that other regulatory mechanisms must be present to enable Keap1-independent Nrf2 activation, possibly by means of posttranslational modifications of both proteins or via interplay with alternate signalling pathways (Z. Zhang et al. 2023; D. Ma et al. 2020)

Moreover, across species, interspecies variation in TP effectiveness and responsiveness to the Keap1/Nrf2 pathway is particularly complex for a number of reasons. These include differences in TP absorption, metabolism, and bioavailability between species, as well as divergent baseline redox states and genetic polymor-

phisms in regulatory elements. The use of experimental variables, such as dietary composition, TP dose and chemical composition, tissue-specific patterns of expression, tissue obesity and controlled chamber conditions, also modulates these. Although dose-dependent effects are common, with low SPI concentrations improving antioxidant defence and immune activity but superbude concentrations could be some physiological exhaust or hepatotoxicity, as shown and in another place mammalian models (Qin et al. 2024; Wei et al. 2023). Overall, although numerous factors influence the Keap1/Nrf2 pathway response, variability in gene expression is likely to be the most common. Thus, it is critical that the genetic variability and other conserved elements are used to aid different expressions. Green TPs (especially EGCG) can increase antioxidant enzymes expression and activity, especially SOD and CAT, through several pathways, such as Nrf2 activation and regulation of gut microbiota. Nonetheless, this antioxidant effect is dose- and species-specific, because high-dose EGCG could be pro-oxidant. The current findings highlight the importance of dosage for the antioxidant effects of TPs and the optimum dose with maximum benefit and minimal side effects. Thus, EGCG and its derivatives are ideal candidates for establishing functional feed additives for enhancing oxidative stress tolerance in aquaculture species.

3.2 | Stress and Disease-Response Genes

Gene families in fish that respond to environmental stressors and pathogens, such as TLRs, play a key role in immune defence. Green TPs can modulate the expression of these genes, enhancing stress tolerance and pathogen defence. Modulation of stress and disease-response genes in fish under green tea feed supplementation is summarized in Table 2. Among the three major gene collection from which the differential genes could be enriched in are heat shock proteins (HSPs), TLRs and some genes of the NF- κ B pathway (M. Qian et al. 2025).

3.2.1 | HSP70

Water temperature is a critical factor influencing fish growth and survival. As ectothermic organisms, fish depend on ambient temperature for metabolism, immune responses and cell homeostasis. Temperature fluctuations in aquaculture can lead to stress, immune suppression and tissue damage (Aboka et al. 2017; H. Y. Yang et al. 2018). A comfort range exists for fish beyond which there is a potential of unrecoverable damage to organs and tissues. The gills (respiratory and immune organs) were affected by high temperature exposure, resulting in structural damage such as aneurysm and lamellar fusion (W. Li et al. 2018; Y. Chen et al. 2021). Meanwhile, heat stress disrupts the cellular redox balance, resulting in the overproduction of ROS, causing lipid peroxidation, cellular atrophy and immunosuppression (Hoseinifar et al. 2020; Tan et al. 2019).

Heat stress induces the production of HSPs, especially HSP70, which help cells cope with thermal stress. These proteins are essential for protein folding, stabilization and repair of damaged proteins during stress conditions (Cardoso et al. 2015). Considering its prompt and persistent expression in reaction to increased temperature we expected that HSP70 could be frequently adopted

TABLE 2 | Modulation of stress and disease-response genes in fish under green tea feed supplementation.

Fish species	Stress/disease condition	Detailed treatment description	Supplementation details	Gene(s)/biomarkers	Expression pattern	Significance	References
Largemouth bass (<i>Micropterus salmoides</i>)	Nutritional supplementation	56-day feeding trial with 4 dietary TP levels (0.00%, 0.02%, 0.04%, 0.08%) plus intermittent feeding group (cycled 7 days TP8/7 days basal diet)	Tea polyphenols (TPs) at 0%–0.08% diet	Antioxidant enzymes: SOD, CAT; immune enzyme ACP; intestinal morphology parameters; microbial composition	Upregulation of SOD, CAT and ACP activities; increased villus height, width, muscular thickness; reduced liver fatty infiltration; decreased abundance of potential pathogens (e.g., <i>Clostridiaceae</i>); improved gut metabolism pathways	$p < 0.05$	Yang et al. (2025)
Stellate sturgeon fingerlings (<i>Acipenser stellatus</i>)	Sublethal B α P toxicity with HSP inducer	Exposed to 100 mg/L Nopal endurance (HSP inducer) for 4 h, followed by exposure to B α P at 25%, 50% and 75% of 96-h LC ₅₀ ; samples taken on Days 1, 3, 6	HSP inducer (Nopal endurance) + B α P at sublethal concentrations	hsp70; CYP450 activity; AChE; SOD; CAT; T-AOC; lysozyme; IgM; C3	Increased <i>hsp70</i> expression at higher HSPi + B α P concentrations; CYP450 activity increased; AChE activity decreased by B α P; antioxidant enzymes and immune markers increased by HSP inducer	Not reported	Vahdatirad et al. (2024)

(Continues)

TABLE 2 | (Continued)

Fish species	Stress/disease condition	Detailed treatment description	Supplementation details	Gene(s)/biomarkers	Expression pattern	Significance	References
Chinese rice field eel (<i>Monopterus albus</i>)	None (nutritional supplementation trial)	9-week feeding trial with isonitrogenous and isolipidic diets containing 0, 25, 50, 100, 200 and 400 mg/kg EGCG	EGCG at 100 mg/kg diet	<i>mhc-2α</i> , <i>hepcidin</i> , <i>il-8</i> (immune genes); <i>il-6</i> , <i>nf-κb</i> (inflammatory genes); AST, CAT, GSH, MDA, LZM, IgM	Upregulation of <i>mhc-2α</i> , <i>hepcidin</i> , <i>il-8</i> ; downregulation of <i>il-6</i> and <i>nf-κb</i> ; decreased AST (except 100 mg/kg), CAT and GSH decreased with dose, MDA increased, increased LZM and IgM	$p < 0.05$	Deng et al. (2023)
Genetically improved farmed tilapia (<i>Oreochromis niloticus</i> , GIFT)	Nutritional supplementation	8-week feeding trial with graded TP levels (0, 100, 200, 300, 400, 500 mg/kg); 15 fish per tank, 6 tanks in quadruplicate	Tea polyphenols (TPs) at 0–500 mg/kg diet	Growth (final body weight), plasma biochemical parameters (LDL, TG), antioxidant biomarkers (CAT, SOD, T-AOC, GSH, MDA)	Reduced final body weight at 500 mg/kg; increased moisture and decreased crude protein at 100 mg/kg; LDL elevated > 200 mg/kg; TG decreased; CAT increased (except 300 mg/kg); SOD increased at 200, 400, 500 mg/kg; T-AOC enhanced at 400–500 mg/kg; GSH improved at 500 mg/kg; MDA elevated at 100–200 mg/kg	$p < 0.05$	Daniel et al. (2024)

(Continues)

TABLE 2 | (Continued)

Fish species	Stress/disease condition	Detailed treatment description	Supplementation details	Gene(s)/biomarkers	Expression pattern	Significance	References
Nile tilapia (<i>Oreochromis niloticus</i>)	Oxidative stress induced by high temperature	70-day feeding trial: 42 days feeding with Gracilaria extract (GE) supplemented diet, then 14 days temperature ramping (1°C every 2 days to 35°C), followed by 14 days holding at 32°C	Red seaweed extract (<i>Gracilaria tenuisipitata</i>), rich in polyphenols	Tight junction genes (<i>occludin</i> , <i>claudin1</i> , <i>ZO-1</i>), <i>nrf2</i> , <i>hsp70</i> ; oxidative stress marker malondialdehyde (MDA); gut microbiota composition	Upregulation of tight junction genes and <i>nrf2</i> ; downregulation of <i>hsp70</i> ; decreased MDA; increased muscle fibre size and RBC counts; improved gut bacterial diversity with increased Firmicutes, Lactobacillales, <i>Sphingobacterium</i> and <i>Prevotella</i> abundance	$p < 0.05$	Siddik et al. (2024)

as a molecular biomarker of thermoregulation in fish. It also plays a role in regulating apoptosis and antioxidant enzyme activities, protecting cellular integrity (Y. Wu et al. 2020; Gandar et al. 2017). For instance, it was reported that HSP70 gene expression was highly upregulated in spotted sea bass (*Lateolabrax maculatus*) in response to increased water temperature, which was associated with histopathological alterations such as detachment and lamellar epithelial curling in the gills—symptoms of heat induced damage to tissue (Phrompanya et al. 2021). Likewise, *Acipenser schrenckii*, *C. carpio* and *Oreochromis niloticus* show elevated respiration rates and increased stress responses to heat, and HSP70 are considered as a major defence response (J. L. Sun et al. 2019; Z. Zhao et al. 2017; Xie et al. 2017; Jia et al. 2020). HSP70 is induced to different extent by heat stress among different species. For example, Fish species with different thermal backgrounds display differential induction of HSP70 in response to temperature stress. An increase (+6°C) in temperature (26°C–32°C) rapidly (< 3 h) and transiently induced the upregulation of multiple HSP70 isoforms in the eurythermal spotted sea bass *L. maculatus*, with expression peaking six- to eightfold above baseline within 3–12 h (Long et al. 2023). This is opposed to cold water, stenothermal brown trout (*Salmo trutta*) which had substantial HSP70 induction only after a larger (+11°C; from 9°C to 20°C) temperature increase, with higher expression maintained over a 21-day exposure period and followed by lower energy reserves and impaired growth suppression (Hampuwu et al. 2025).

Whilst warm-water species may be mounting rapid yet brief responses in HSP70 expression to smaller thermal challenges, cold-water species are requiring larger temperature changes before a prolonged induction of HSP70 expression which is indicative of their limited thermal tolerance. This is crucial in both determining species vulnerability to temperature stress, and in managing temperatures in aquaculture to cope with an increasingly erratic climate. Specific HSP70 isoforms like HSPa1a have also been reported to be involved in thermotolerance in studies. HSPa1a regulates autophagy and cell survival signalling and provides cells with tolerance to heat stress (Y. Wu et al. 2020). Furthermore, bacterial infection and temperature stress both cause rainbow trout (*O. mykiss*) and channel catfish (*Ictalurus punctatus*) to express HSP70, suggesting that HSP70 serves two purposes in immunological and thermostress reduction (Li, Yu, et al. 2022; L. Sun et al. 2016). In Japanese flounder (*Paralichthys olivaceus*), five out of the fifteen HSP70 genes were upregulated after *Edwardsiella tarda* infection, thus further supporting the role of HSP70 in the host immune response (Jiang et al. 2020).

The genome-wide discovery of members of HSP70s family has been reported in various aquaculture species. In contrast, at least 17, 20 and 20 HSP70 isoforms were identified in spotted seabass (*Larimichthys crocea*), large yellow croaker and mudskipper (*Boleophthalmus pectinirostris*) upon environmental stress, respectively (L. Liu et al. 2020; Y. Chen et al. 2021). In turbot (*Scophthalmus maximus*), which is extremely sensitive to environmental variations, phylogenetic and transcriptomic analyses identified and systematically analysed, several HSP70 genes and demonstrated that these HSP70 genes had regulatory functions during heat, salinity and pathogenic stress (J. L. Sun et al. 2019; X. Li et al. 2020; F. Zhao et al. 2021; Y. Liu et al. 2021). Notably, dietary intervention has become one of the successful means of

manipulating HSP70 expression in fish. Hepatic and gill HSP70 in fish such as *O. niloticus* and *C. carpio* display upregulation following exposure to green TPs such as EGCG concurrent with increased antioxidant enzyme activities and attenuation of oxidative stress levels (Hasanpour et al. 2017). These findings show that the protective effects of green tea bioactives may be achieved in part by activating HSP70 and controlling ROS.

Taken together, HSP70 is not only a molecular chaperone, but also a genomic marker for stress tolerance and immune resistance for fish. Its expression is closely related to extrinsic stimuli and to diet control. The addition of green tea extracts (GTEs) in aquafeed presents itself as a potential approach to promoting fish health by triggering cellular defence mechanisms, especially through HSP70-dependent mechanisms. This synergistic mechanism may vary across species and requires further research to clarify its specific role in aquatic animals. To identify a complete repertoire of R and NR genes, and to improve functional feeds strategies in sustainable aquaculture, further studies using transcriptomics, promoter activity mapping, as well as epigenetic regulation are likely required.

3.2.2 | TLRs

TLRs are key receptors involved in the innate immune response to microbial pathogens in aquaculture species. TLR3 and TLR4, in particular, are important for antiviral and antibacterial defence, especially under stress conditions like heat (Y. Li et al. 2023; Kong et al. 2023).

3.2.2.1 | TLR3 in Teleosts: A Sentinel Against Viral and Bacterial Threats. In common carp (*C. carpio*), people have demonstrated increased poly I:C-induced expression of an antiviral gene Mx in several organs, including the kidney, liver and spleen after ip injection of the double-stranded RNA (dsRNA)-mimic, poly I:C. Likewise, poly I:C upregulates the expression of interferon regulatory factor 3 (IRF3), a pivotal transcription factor downstream TLR3, in the orange-spotted grouper's spleen (*Epinephelus coioides*). In the immune tissues of Japanese sea perch (*Lateolabrax japonicus*), TLR3 had high expression level and significantly elevation after bacterial challenge, indicating a wide innate immune function (Wang, Zhao, et al. 2018). TLR3 in golden pompano (*Trachinotus ovatus*) exhibited similar expression patterns except that there were tissue-specific and time-dependent responses in head kidney, spleen and liver (S. Wu et al. 2021). The induction of the *TLR3* gene in megalopae larvae after exposure to poly I:C is indicative of its function in dsRNA recognition and stimulating immune response (Q. N. Liu et al. 2016; R. Zhu et al. 2017) in yellow catfish (*Pelteobagrus fulvidraco*). However, the molecular basis of TLR3 signalling in teleosts is still not well understood. Nevertheless, advancement of new discoveries is now possible due to developing bioinformatics tools and genomic resources. For example, Alpha Fold, a deep learning model of protein structure prediction, can help to characterize the spatial arrangements of immune proteins (Ikeya et al. 2019; Jumper et al. 2021).

Comparative genomics assisted by chromosomal level assemblies such as that of yellow catfish (Gong et al. 2018) facilitates recognition of homologous TLR genes among species and can

provide estimates of the rate of evolution by calculating the dN/dS ratio (Spielman and Wilke 2015). Toll-like receptor 3 (TLR3) is an intracellular PRR that detects dsRNA of viral viruses via interferon regulatory factors and pro-inflammatory cytokines leading to antiviral immune response which signals through downstream cascades (Beutler 2002; Kawai and Akira 2010). In Cyprinid fish, intraperitoneal injection poly(I:C) in common carp (*C. carpio*) causes a known increase of the antiviral Mx gene in spleen, liver and kidney (Falco et al. 2014), also characterized by rapid response to viruses through a peak of TLR3 transcription at only 12 h with cyprinid herpesvirus 2 infection in Prussian carp (*Carassius auratus gibelio*). Primary antiviral, TLR3 is also involved in response to bacterial challenges; with transcription in spleen and kidney of hybrid yellow catfish (*P. fullvidraco* × *P. vachelli*) obviously elevated upon infection by *Aeromonas hydrophila*, but apparent at different timepoints compared to poly(I:C) stimulation (Q. N. Liu et al. 2016). However, a comparative phylogenetic analysis revealed that TLR3 was conserved from vertebrate to invertebrates since TLR3-like genes were annotated in crustaceans such as the whiteleg shrimp (*Litopenaeus vannamei*), which play important roles in antiviral defence against dsRNA viruses (Guo et al. 2021). This indicates that it has played an ancient and essential role in pathogen perception across aquatic taxa.

All things considered, TLR3 serves a conserved role in the recognition of viral and bacterial components in fish, and the insight into its regulation and modulation—through dietary approaches such as green TPs—opens new perspective for strengthening aquaculture immunity.

3.2.2.2 | TLR4 in Bivalves: Environmental Stress and Innate defence. Ocean warming is a major threat for marine bivalves, which are poikilothermic and sessile animals. Immune responses of bivalves can be influenced by acute heat stress, and this reaction is linked to TLR4. According to Saco et al. (2023), bivalve TLRs have the more variety of members in animals, which has possibly resulted from their adaptation to diverse environmental niches challenged by pathogens.

Bivalve TLRs are not orthologous to those of vertebrates, but evidence is accumulating of the existence of a functional TLR4–MyD88 pathway (Xing et al. 2017). Eighteen TLR genes were found in Yesso scallops (*Patinopecten yessoensis*), among which TLR4 was found to be inducibly expressed under acidification stress. The same result was found in *C. hongkongensis* and the combined results of transcriptomic studies and the RNAi assay proved that TLR4 acted in immune defence against *Vibrio parahaemolyticus* (VP) (Chen, Gao, et al. 2022; F. Yu et al. 2023). The expression of TLR4 was also associated with immune responses in *Pinctada fucata martensii* after implanting no nucleus, a stressor often found in pearl production (Lu et al. 2022). Further characterization of full-length cDNAs of HMTLR4, HMMyD88 and HMTRAF6 proved the existence of a TLR4–MyD88 cascade. Therefore, the expression of a subset of these genes was modulated under thermal stress, and/or LPS challenge, being repressed in general at over 30°C contrary to a pro-inflammatory activation in homeotherms, such as pigs (X. H. Ju et al. 2014; Cantet et al. 2021; Martine and Rébé 2019). Despite the gene suppression, levels of TNF- α and LZM were considerably enlarged in LPS-injected scallops, indicating the presence of

alternative or complementary immune pathways. Furthermore, RNAi against HMTLR4 also resulted in a reduction of the downstream effectors' abundance, confirming the involvement of the HMTLR-mediated cascades in bivalve innate immunity (F. Yu et al. 2023; Parisi et al. 2022). These outcomes indicated that thermal stress has affected the TLR4 signalling in bivalve, which might be related to the participation of HSPs, similar with the observation in mammals that HSP70 induces a decrease of TLR4 in vivo (Ghosh et al. 2015). Here it is found that HSP70 plays a context-dependent dual role in modulating the TLR/NF- κ B signalling pathway. Extracellular HSP70, which is passively released by injured cells becomes a damage-associated molecular pattern (DAMP) capable of activating TLR2 and TLR4 to induce pro-inflammatory responses through NF- κ B signalling. In chronic or severe heat stress, high level intracellular HSP70 inhibits TLR4 expression to reduce downstream NF- κ B signalling and avoid excessive inflammation situation and tissue injury. Therefore, this feedback regulation serves to counterbalance between immune activation and cytoprotection (Guo et al. 2023).

In addition, genes in the TLR4–MyD88 pathway were downregulated by acute heat stress (> 30°C) in marine bivalves such as Hongmo no. 1 scallops, which is contrary to the canonical pro-inflammatory response typically observed in homeotherms. Such suppression probably reflects an adaptive modulating mechanism to prevent excess in hyperinflammation induced by thermal stress. Importantly, HSP70 suppresses TLR4 availability or adaptor protein interaction as well as NF- κ B stimulation possibly through the direct inhibition analogously to mammalian models (C. Yue et al. 2024). Together, these findings demonstrate an evolutionarily conserved synergistic mechanism for HSP70 fine-tuning of TLR/NF- κ B signalling to regulate the innate immune response and stress tolerance in mammals. It is important to know the regulation of the TLR4 signalling pathway under temperature stress for process of disease resistance in aquaculture. There is potential for dietary polyphenols, including green TPs, for stabilizing or reinvigorating TLR signalling under stressful conditions as a furious area of future research.

3.2.3 | NF- κ B Pathway Activation

The NF- κ B signalling pathway influences intestinal barrier function, inflammation and immunity in fish and shrimp. Green TPs, especially EGCG, are being explored as potential regulators of this pathway in aquaculture species (Deng et al. 2023). TPs in the diet have been reported to mitigate LPS-induced intestine damage via the regulation of TLR2/MyD88-mediated NF- κ B activation in *C. carpio* (Gao et al. 2019). TPs repressed the expression of pro-inflammatory cytokines IL-1 β , IL-6 and TNF- α , DNPs of NF- κ B (Barnabei et al. 2021; Wang et al. 2022). Molecular docking confirmed such action mode and showed good binding between TPs and TLR2b and substantial inhibition over the bacteria ligands recognition sites (NF- κ B activation). This mechanistic view is in agreement with the interaction model shown in Figure 1 (Shen et al. 2017; Mohammadi et al. 2022). Moreover, TPs notably suppressed the activity of the NF- κ B/MLCK pathway, which is an important mediator of tight junction modulating. However, the impaired phosphorylation of MLC (p-MLC) was determined as a result of the blocking of MLCK's activity; it was showed that

tight junction proteins contain occludin, claudins and ZO-1 were preserved (J. Zhong et al. 2021; S. Ma et al. 2023; S. Chen et al. 2015). Histologically, the above molecular effects were verified by improved intestinal tissue architecture, lessened epithelial injury and improved mucosal barrier function in TP-fed fish (Yuan et al. 2021). The anti-inflammatory action of TPs was not the only immunomodulatory effects. Increased expression of mucosal immune factors LZM, alkaline phosphatase (ALP), immune globulins (IgM, IgD and IgT) and complement proteins (C3 and C4) has been noticed in various other teleosts on green tea supplementation confirming activation of both innate and adaptive immune responses (Debbarma et al. 2022; Espinosa et al. 2020). Green TPs offer multi-pronged prevention against SI-ARTI by targeting TLR2–MyD88–NF- κ B signalling in an omics-based approach. This enhanced immunology, epithelial barrier and inflammation regulation. These molecular findings indicate that green tea can be utilized as a functional feed additive for enhancing the fish aquaculture disease resistance and stress tolerance.

4 | Green Tea as a Natural Alternative to Antibiotics

4.1 | Ecological and Health Implications of Antibiotic Use in Aquaculture

More recently, attention has been placed on the ecological and health effects of antibiotics in aquaculture. The overuse of antibiotics has created resistance, which is a threat to human health and the environment (Okeke et al. 2022; Lulijwa et al. 2020; Nandi et al. 2025).

From an ecological perspective, the extensive use of antibiotics in aquaculture leads to the emergence of resistant bacterial strains, which might be transferred to humans due to feeding and environmental exposure (Maixent et al. 2023; Hridoy, Akter, et al. 2025). Research has shown that moving towards novel interventions is essential for a sustainable aquaculture. Green tea, which is abundant in polyphenols and most notably EGCG (epigallocatechin-3-gallate) exhibits antimicrobial activity against waterborne aquatic pathogens in environments (Li et al. 2022; Sheikhzadeh et al. 2011; F. Zhao et al. 2021). These properties highlight its role in the prevention of fish diseases and thus it reduces dependency on synthetic antibiotics that have been ban or stringently control in many regions (Chakraborty et al. 2014). Botanicals used in dietary supplements, nutraceuticals and functional foods is; GTE. TILAPIA, is a warm water fish and is a popularly farmed fish and research has proved that frozen GTE preserved fish has more shelf life than ice. In addition, addition of green tea in the diet of animals has resulted in the improved health and survival in fish and in aquatic hardships conditions (Y. Chen et al. 2019).

The health benefits of feeding green tea to cultured fish go beyond pathogen suppression. The immune modulatory effects of green tea derivatives and its role in enhanced resistance against different aquatic species have been evidenced. For example, findings have shown that green tea can improve growth rates and support the immune system of fish, and thus, it has a bivalent role in growth promotion and protection (Abo El-Maaty et al.

2023; Abdel-Tawwab et al. 2010). The benefits of tea are mainly attributed to its function as an immune stimulant in various animals including fish (Sheikhzadeh et al. 2011). GTE supplementation stimulated the lipolytic pathway by enhancing the activities of hormone sensitive lipase (HSL) and comparative gene identification-58 (ABHD5) and this was reflective by decreased body weight and adipose tissue gain. Furthermore, green tea modulated the expression of critical regulators of glucose transport and insulin signal pathways, leading to whole-body lipid concentration decrease (Cao et al. 2007).

Being an environment-friendly additive, green tea is safe and eco-friendly compared to the commercial synthetic additives (Y. Chen et al. 2019; Hridoy, Shawkat, et al. 2025; Hridoy, Dittthakit, et al. 2025; Lima et al. 2025). Its inclusion in the diet of livestock can enhance feed efficiency and gut health through manipulation of the gut microbiome (Y. Chen et al. 2019). Furthermore, green tea might enhance nutrient utilization thus improving feed conversion and having decrease faecal excretion of waste (Gulua et al. 2024). By contrast, artificial additives, as they contain chemical substances, have negative consequences on the natural environment and consumer and residual adverse effects. On the other hand, green tea has been reported as safe for the animals receiving it and for the humans using such products (Thomas et al. 2022).

Using green as a non-toxic antibiotic in fish represents many ecological and health advantages. It tackles pressing issues on antibiotic resistance and promotes sustainable development through boosting the immune systems of aquatics and promoting the animal husbandry system. Further studies on its application could make green tea an available and efficient ingredient in aquaculture feeding programs.

4.2 | Benefits of Green Tea as an Eco-Friendly Additive

While green tea offers an eco-friendly alternative to antibiotics, its effectiveness as a disease-preventative feed additive is still debated due to inconsistent results across species and environmental conditions. Further studies are needed to evaluate its long-term benefits and potential drawbacks in aquaculture systems (Kumar et al. 2023., Singh et al. 2022). The non-toxic feature of green tea is one of the most important benefits to use it in aquaculture. In contrast to some chemical additives which are synthetic and may introduce harmful substances into the aquatic environment with the possible subsequent accumulation of these substances into the wide food chain, green tea is viewed as having a low-risk potential for both aquatic organisms and consumers. Polyphenolic contents of green tea manifest antimicrobial activities against pathogenic bacteria and are likely to be a safer option than antimicrobials due to the absence of harmful residues (Zorriehzahra et al. 2016; N. Zhang et al. 2025). This feature serves to not only protect the health of fish populations, but also to foster a healthy ecosystem by lowering the burden of pollutants in the environment (Zorriehzahra et al. 2016., Rodrigues et al. 2024; Hridoy, Bordin, et al. 2025; Hridoy, Neogi, et al. 2025).

Moreover, green tea is one of the sustainable source of food and is grown under minimal use of synthetic pesticides and fertilizers; thus, it is considered as greener food for aquaculture feed (Kausar 2021; Hridoy et al. 2021). It is produced jointly with eco-friendly farming, which is gaining popularity as customers becomes increasingly aware of food production systems being harmful to the environment. The eco-benefits of green tea as an additive can render it desirable to incorporate, which is suggestive for a greater sustainable management in fish farming (Thomas et al. 2022).

The immune strengthening qualities of green tea are especially remarkable. GTEs are also able to improve resistance against diseases in cultured fish (Thomas et al. 2022). This immune-modulatory action can enhance the growth and general performance of the aquatic species with consequences for the increased yield in aquaculture. However, the use of synthetic additives, aside from being effective for some purposes, can alter the natural resistance of fish through their ability to modulate functions and activities of the microbiota of the gut, which may lead to dysbiosis (Zorriehzahra et al. 2016).

In comparison with synthetic enhancers commonly applied in the diets for aquaculture, green tea shines as an excellent substitute. Synthetic additives typically have many side effects with possible toxicity and environmental hazards. For example, overutilization of antibiotics can result in the promotion of resistant bacteria, leading to use restrictions by some regulatory agencies (Song et al. 2017). This is a response to invasive chemicals and compounds pushing forwards with little regard for FPSs, but is at the same time a 'push back' to the business as usual, since green tea as 'nature's drugstore' is a support system in many directions for aquaculture, related to health, welfare, growth and sustainability and without the danger of side effects from synthetic choices. Currently, there is no significant data available to suggest that green tea contributes to environmental toxicity; however, future studies are needed to explore this aspect further.

5 | Omics-Based Evidence of Green Tea's Effect on Fish and Shrimp

Green tea is gaining attention as a feed ingredient in aquaculture due to its positive effects on health, growth and disease resistance. Its bioactive components, such as L-theanine, caffeine and catechins, influence various physiological processes, including immune regulation and stress adaptation, as revealed through advances in transcriptomics and genomics (Y. C. Qian et al. 2021). One of the most significant successes is the identification of key pathways affected by green tea supplementation in aquaculture species using omics technologies: transcriptomics and qPCR. Research has shown that green tea affects immunological, metabolic and oxidative gene expression, focusing on how it mitigates the negative effects of long-term stress in fish and shrimp (Sánchez-Velázquez et al. 2024; Zeng et al. 2020; Liao et al. 2022).

However, it has been shown that the bioactive components of green tea influence the host's responses genetically through gene expression analyses. It has been found that green tea strengthens overall physiological resilience, increases immuno-

logical systems and fosters pathogen resistance in agitated fish and shrimp. Green tea's implications for growth-related genes, immunological markers and metabolic pathways important to stress adjustment have been determined by scientists using high-throughput transcriptome techniques. These molecular insights provide a rational basis for the specific application of green tea, as a functional dietary supplement able to enhance fish health, disease resistance and production efficiency in intensive aquaculture (Zhao, Liang, et al. 2022; W. Li et al. 2018). The application of omics technologies in aquaculture has expanded our understanding of how bioactive green tea compounds reduce stress tolerance in aquatic life. Studies using qPCR and RNA sequencing confirm that green tea supplementation has the potential to positively impact antioxidant pathways by increasing enzymatic defence capacity while against reactivate of ROS accumulation in conditions of environmental stress such as poor water quality or disease outbreaks. The alteration of crucial genes involved in stress response pathways, particularly those connected with oxidative stress and immunological defence, emphasizes how well green tea increases the resilience of farmed aquatic species (Huang et al. 2023).

The technical and practical constraints must be considered when interpreting omics-based evidence for green tea efficacy in aquaculture, although promising results have been observed. Most feeding trials are short-term (e.g., for up to 6 weeks) which constrains researchers' ability to assess long-term physiological adaptations, growth performance and response traits over full grow-out cycles in a commercial type setting (J. L. Chen et al. 2011). Responses are highly species-specific also, for example, whereas *Nile tilapia* and red sea bream display clearly improved growth and anti-oxidative status with oxidised oil and GTE in the diet, response of sturgeon hybrids was little sensitive to dietary GTE or oxidised oil which might be due to palatability/make-up of its metabolism (Tocher et al. 2003; Gao et al. 2012). Second, anti-nutritional effects can also be induced by excessive dietary inclusion of TPs. For instance, in large yellow croaker, Ji et al. (2018) found that lipid oxidation and lipolysis genes including acyl-CoA oxidase (ACO), carnitine palmitoyltransferase-1 (CPT1) and peroxisome proliferator-activated receptor alpha (PPAR α) were significantly downregulated by 0.05% TP inclusion compared to lower concentrations, implying the metabolic regulation beyond certain levels are lost in such condition. Furthermore, the beneficial effects of GTE may be diet- or species-dependent; while oxidative stress and lipid metabolisms were disturbed when 25% of fish meal was replaced by soybean meal in largemouth bass, supplementation with plant-based functional additive containing GTE ameliorated these disturbances and it might not have similar results under different baseline diets or species scales (Zhou, et al. 2023). Third, experimental stress models typically involve acute stresses, for example, 24-h heat shocks to mimic heatwave conditions associated with climate change. Though informative, this model is not appropriate for studying chronic adaptation processes that may include the sustained expression of HSPs or slow shifting of energy metabolism (N. Zhang et al. 2025). In oxidative stress evaluations, the use of MDA as the most important lipid peroxidation marker is limited by ignoring other aldehydic products, such as 4-hydroxynonenal (4-HNE) adducts, which detrimental effects cannot be explained only by MDA and provides a better picture about lipid damage (Faizan et al. 2013; Brigelius-Flohé and Maiorino 2013). Although TPs have been

proposed to restore antioxidant enzyme activity by both direct ROS scavenging and Nrf2-mediated transcriptional activation in fish (L. Zhong et al. 2020; Hu et al. 2019), the precise molecular mediators are currently unknown, warranting targeted lipidomic and transcriptomic verification.

Last, TPs are inherently unstable in biological systems and will rapidly degrade at physiological temperature, O₂ conditionality and metal ion exposure, yet more so in the neutral to alkaline pH upshot. These characteristics make it an unstable compound, with low oral bioavailability and a short plasma half-life after oral administration (J. Jin et al. 2022; Krupkova et al. 2016; Xu et al. 2019; Dang et al. 2013). The stability can be improved with encapsulation in, for example, zein-Gum Arabic complex nanoparticles but the antioxidant potency of the probiotics can also decrease because antioxidants activity is influenced by hydrogen bonding and other interactions with carrier matrices (Y. Li et al. 2009; Gülcin 2020). Moreover, differences in nanoparticle formulations and the inclusion of secondary stabilizers hamper reproducibility and meta-analytic synthesis. Resolving these technical limitations will be key to taking the promising molecular advantages of green tea ingestion to scalable, reproducible solutions for supporting sustainable aquaculture.

5.1 | Quantitative PCR and Transcriptomics Evidence

While qPCR and transcriptome analysis have helped identify key pathways influenced by green tea supplementation, conflicting results across species and experimental conditions raise questions about the generalizability of these findings. The variability in gene expression and the influence of external factors like diet composition and baseline health highlight the need for more consistent study designs and larger-scale trials (Liao et al. 2022; Zeng et al. 2019; Power et al. 2023). The qPCR proves to be a sensitive method used for the quantification of gene expression, particularly of genes involved in immunity, anti-oxidative defences and inflammation. It has been shown in different studies that immune associated genes (i.e., interferon, *IL-1 β* , *TNF- α* , LZM and C-reactive protein) are commonly modulated by green tea in fish species, such as Nile tilapia (*O. niloticus*), gilthead seabream (*Sparus aurata*) and gibel carp (*Carassius gibelio*) (S. Li et al. 2021; Elabd et al. 2022; Teixeira et al. 2022). These differential gene expressions in response may contribute to enhanced immune function, increased disease resistance and greater survival under pathogenic stress.

In addition, transcriptomic studies, especially those based on RNA sequencing (RNA-Seq), have given a global picture of gene transcription networks in fish fed green tea. The DEGs of fish including rainbow trout (*O. mykiss*) and common carp (*C. carpio*) were enriched in cytokine signalling pathways, regulation of oxidative stress response, heat shock and pathogen recognition pathways (Nootash et al. 2013; G. Yang et al. 2022). Specifically, the upregulation of genes involved in the TLR and NF- κ B pathways is consistent with the proposed beneficial effect of green tea bio-actives on innate immune and inflammatory modulations (Q. Li et al. 2019; G. Yang et al. 2022). Transcriptomic analysis of gene expression in fish and shrimp fed green tea is shown in Table 3. Moreover, transcriptomics have shown that green

tea compounds, in particular, TPs and EGCG, have an impact on central metabolic and stress pathways. More importantly, green tea has demonstrated to mediate the AKT/AMPK/mTOR signalling pathway as well as H₂O₂-induced oxidative stress response that is important for maintaining cellular homeostasis in environment and nutritional stress (C. S. Yang and Hong 2013; Tian et al. 2022; Truong and Jeong 2022). These changes are generally associated with upregulation of antioxidant enzyme genes (SOD, CAT and glutathione peroxidase [GSH-Px]) and the downregulation of pro-inflammatory factors (IL-8 and IL-1 β) (Z. F. Yang et al. 2014; Liu, Wang, et al. 2023).

Combined transcriptome data of qPCR studies indicated that green tea has potential to be an immune and stress-relieving feed additive in aquaculture. These resources do not only disclose species-specific transcriptional responses but also indicate the wide potential of green tea to enhance fish health and resistance to diseases by action on pivotal physiological and cellular pathways (Liao et al. 2022; Power et al. 2023; Q. Li et al. 2019).

5.1.1 | Impact on Fish Growth and Immunity

Green tea supplementation can regulate immune, antioxidant and metabolic gene expression, but conflicting results in terms of growth and immunity across species require more standardized trial conditions. While qPCR studies indicate upregulation of immune-related genes like IL-1 β and *TNF- α* in some species, these effects are not universal across all aquaculture species (Elabd et al. 2022; Wang et al. 2021). These alterations are associated with enhanced immune response resistance to seminal pathogens in fish species such as Nile tilapia (*O. niloticus*), gilt-head seabream (*S. aurata*) and gibel carp (*C. gibelio*) (Wang et al. 2021). This is supported by transcriptomic studies, which report differentially expressed genes (DEGs) implicated in immune signalling, pathogen recognition and regulation of inflammation. For example, GTE can influence genes related to cytokine signalling, TLR pathways, as well as the NF- κ B cascade, all of which are crucial elements of the innate immune system (R. Zhang et al. 2020). These effects result in a visible phenotypic enhancement including improved disease resistance, survival and decreased pathogen load upon infection challenge (S. Li et al. 2021; Wang et al. 2021). Green tea, apart from having an immunomodulatory role, has been shown to be antioxidant and stress alleviating through qPCR and transcriptomic studies where the expression of antioxidant enzyme genes such as SOD, CAT and Nrf2 is upregulated (S. Li et al. 2021; Q. Li et al. 2019; Tian et al. 2022). These genes are important for scavenging of ROS and promotion of cellular defence.

Furthermore, GTE supplementation has also been associated with reduced expression of pro-inflammatory cytokines, including IL-8 and IL-1 β , highlighting its anti-inflammatory properties (S. Li et al. 2021; Wang et al. 2021). Metabolism of genes regulation of genes is another key effect of green tea supplementation. Results of RNA-seq and qPCR analyses indicated that green tea bioactive modulate genes involved in lipid and carbohydrate metabolism, muscle development and nutrient uptake (Wang et al. 2021; D. Yue et al. 2021). In grass carp (*Ctenopharyngodon idella*) and hybrid groupers, the addition of dietary TPs was

TABLE 3 | Transcriptomic analysis of gene expression in fish and shrimp fed green tea.

Species	Treatment and duration	method used	Key genes affected	Biological pathways impacted	Metabolomics	References
Chinese rice field eel (<i>Monopterus albus</i>)	EGCG at 0, 25, 50, 100, 200 and 400 mg/kg in isonitrogenous and isolipidic diets for 9 weeks	qPCR; growth performance measurement; biochemical assays (AST, CAT, GSH, MDA, LZM, IgM)	<i>mhc-2α</i> , <i>hepcidin</i> , <i>il-8</i> ↑; <i>il-6</i> , <i>nf-κb</i> ↓	Non-specific immune response enhancement, antimicrobial peptide production, inflammation regulation	Lower whole-body lipid content (25, 50, 400 mg/kg); decreased serum AST (except 100 mg/kg); decreased hepatic CAT, GSH, increased MDA with EGCG level; increased LZM activity and IgM content; linear relationship between EGCG level and immune/oxidative parameters; optimal EGCG ≈ 109.81 mg/kg (IgM)	Deng et al. (2023)
Largemouth bass (<i>Micropterus salmoides</i>)	Tea polyphenols (TP) at 0.00%, 0.02%, 0.04%, 0.08% for 56 days; intermittent feeding group (0.08% TP for 7 days followed by basal diet for 7 days, repeated until day 56)	qPCR; antioxidant and immune enzyme assays; liver and intestinal histology; intestinal microbiota analysis	<i>CAT</i> , <i>SOD1</i> , <i>Keap1</i> , <i>Nrf2</i> , <i>TOR</i> ↑; <i>IL-1β</i> ↓	Antioxidation (Nrf2–Keap1 signalling), immune response regulation, intestinal health, metabolism (amino acid, carbohydrate, lipid)	Reduced crude lipid, increased crude protein; decreased serum CHO and LDL; increased SOD, CAT, T-AOC; decreased MDA; improved ACP; reduced liver fatty infiltration; improved VH, VW, MT, LPS activity; decreased Clostridiaceae; increased <i>Cetobacterium</i> ; altered metabolic pathways	Yang et al. (2025)
<i>Penaeus vannamei</i> (whiteleg shrimp)	Oral feeding of hydrolysable tannin for 28 days (0, 1, 2, 4, 8 g/kg feed); <i>Vibrio alginolyticus</i> challenge on Day 7	In vitro assays, in vivo feeding trial, qPCR	↑ <i>Penaeidin-2</i> , <i>penaeidin-3</i> , <i>penaeidin-4</i> , <i>crustin</i> , <i>lysozyme</i> ; ↑ <i>THC</i> , <i>phagocytosis</i> , <i>respiratory burst</i> , <i>PO activity</i> (especially in T2 and T4)	Immune stimulation, antimicrobial peptide expression, intestinal integrity, host–pathogen resistance	↑ Survival rate post-infection: T2 (66.67%), T4 (69.70%); ↑ intestinal wall thickness; enhanced nutrient absorption and growth performance; optimal dose = 4 g/kg	H. T. Huang et al. (2024)

(Continues)

TABLE 3 | (Continued)

Species	Treatment and duration	method used	Key genes affected	Biological pathways impacted	Metabolomics	References
Hybrid grouper (<i>Epinephelus fuscoguttatus</i> ♀ × <i>E. lanceolatus</i> ♂)	Tea polyphenols (TP) at 0, 0.01, 0.02, 0.04, 0.08, 0.16% (TP1–TP6) in high-lipid diets (51% protein, 16.7% lipid) for 8 weeks	qPCR; antioxidant and immune enzyme assays; histological analysis; <i>Vibrio harveyi</i> challenge test	Liver: <i>sod</i> , <i>cat</i> , <i>gsh-px</i> , <i>nrf2</i> , <i>keap1</i> , <i>hsp70</i> , <i>hsp90</i> , <i>myd88</i> , <i>tnfα</i> ↑; <i>tlr22</i> , <i>il8</i> , <i>il1β</i> , <i>il10</i> ↓. Head kidney: <i>myd88</i> , <i>il1β</i> , <i>tnfα</i> , <i>il6</i> ↑; <i>tlr22</i> , <i>il10</i> ↓	Antioxidant defence (Nrf2–Keap1 pathway), heat shock response, immune regulation via TLR–MyD88 signalling, cytokine-mediated immunity, disease resistance	Increased serum CAT, GSH-Px, T-AOC and liver SOD, CAT, GSH-Px, T-AOC; decreased ROS and MDA; improved ACP, AKP, LYS, IgM; alleviated hepatic pathology; improved survival after <i>V. harveyi</i> challenge; optimal TP ≈ 0.092%–0.106%	Pan et al. (2022)
Zebrafish (<i>Danio rerio</i>)	GTE: 100 µg/mL (larvae, short-term); 250 µg/g BW/day (adult, oral)	RNA-seq, qPCR	↑ AMPK, ↓ SOX9, ↓ MMP2, ↓ SREBF1	WNT/β-catenin signalling, AMPK activation, lipid metabolism, adipogenesis inhibition	↓ Plasma TG and TCHO, ↓ VAT, downregulation of de novo lipogenesis genes, ↑ lipid oxidation genes	Zang et al. (2019)
Penaeid shrimp (<i>P. vannamei</i>)	Clinical samples (<i>n</i> = 190) from multiple Chinese provinces (Shandong, Jiangsu, Sichuan, Guangdong, Hainan)	Triple Eva Green real-time PCR assay (melting curve-based)	Not applicable (pathogen detection)	Accurate simultaneous detection of EHP, IHNV and DIV1; Tm differentiation (EHP: 76.0°C–77.2°C, IHNV: 79.2°C–80.2°C, DIV1: 81.2°C–82.0°C)	Detection rates: EHP (10.5%), IHNV (18.9%), DIV1 (44.2%); effective for early and low-level infections; co-infection detected	Dong et al. (2024)
Exopalaemon carinicauda and <i>Litopenaeus vannamei</i>	<i>Vibrio parahaemolyticus</i> (VP) infection (4 h); quercetin dietary treatment (0.1, 1.0, 2.0 g/100 g feed)	Transcriptomics, metabolomics, qPCR	Toll-like receptor, crustin, AP-1 ↑; DEG downregulated more in low-resistance group (TH04) than high-resistance (Mx)	NF-κB signalling, flavonoid biosynthesis, immune activation, antibacterial defence	11 flavonoids (notably quercetin) ↑ in resistant Mx group; quercetin optimized at 1 g/100 g feed improves immunity and survival; ↑ ACP, LZM, IgM; enriched flavonoid–antibacterial pathways	Jin et al. (2024)

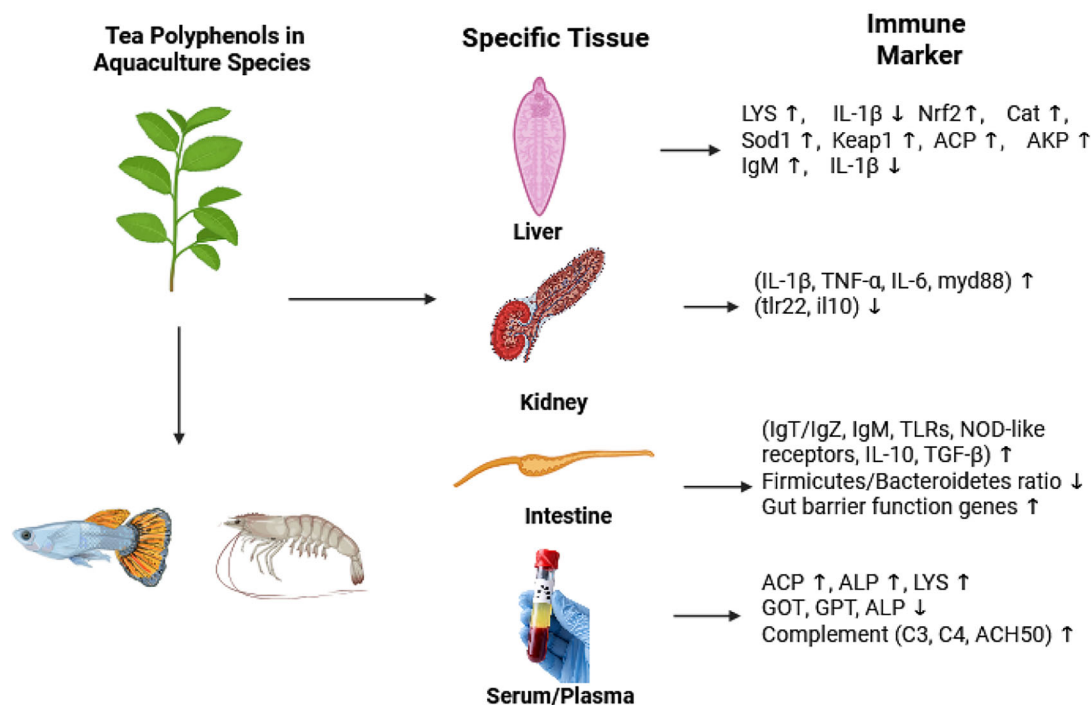


FIGURE 4 | Tissue-specific modulation of immune markers in aquaculture species by green tea supplementation.

beneficial for growth performance, muscle quality and energy utilization pathways in two studies (R. Zhang et al. 2020). However, investigations have also pointed to a role for EGCG in modulating critical signalling pathways, including AKT/AMPK/mTOR and H₂O₂ signalling associated with energy balance, autophagy suppression and protection against ROS (Cheng et al. 2022; Wang et al. 2022). Reproductive growth patterns of various aquaculture species (e.g., coho salmon, rainbow trout) post ingestion of green TPs. In Figure 4, data show dose-dependent weight gain and growth rate as a function of exposure in weeks. The evidence based on transcriptomic and qPCR data demonstrating the differential regulation of immune components, for example, TNF-α, IL-1β, NF-κB, LZM and antioxidant enzymes (SOD, CAT, GSH-Px) among the tissues (liver, serum, head kidney, intestine) in fish and shrimp fed GTEs or polyphenols is presented in Figure 5. These values were obtained from these sources (Yang et al. 2025; Pan et al. 2022; Bilgin, Karabulut, and Tırlı 2025; Daniel et al. 2024; H. Yu et al. 2024).

In mud crab (*Scylla paramamosain*), Kuruma shrimp (*M. japonicus*) and zebrafish (*Danio rerio*), the EGCG treatment stimulated immune gene expression and increased the resistance to viral and bacterial infection (such as *Vibrio alginolyticus* and white spot syndrome virus) (Liu, Wang, et al. 2023, Imperatore et al. 2023).

Similar immune, stress and metabolic parameters including TNF-α, IL-1β, NF-κB, SOD and CAT were uniformly regulated by green tea in different fish species and the prevalent molecular evidence suggests that green tea could be a potential functional feed additive in the field of aquaculture as well. These molecular-level responses were matched with advances at the phenotypic level, such as an increase in growth rate, immune responsiveness, stress tolerance and survival capacity under adverse conditions (Q. Li et al. 2019). Integrating the results of qPCR and transcriptomic

analyses, we conclude that green tea and, specifically, its polyphenolic compound EGCG are indispensable to the health benefits to fish. By modulating the transcriptional reprogramming of immune, antioxidant, inflammatory and metabolic pathways, green tea is a valuable natural solution for improving aquaculture productivity and sustainability (Zeng et al. 2019; Power et al. 2023).

5.1.2 | Effect on Physiological Responses in Shrimp

While green tea flavonoids and polyphenols have been shown to enhance shrimp health, the dose-dependency and species-specific responses to these compounds suggest that more research is needed to determine optimal concentrations for improving immune defence and oxidative stress resistance. Studies indicate that quercetin, the major flavonoid in green tea, has potential immune-enhancing and antioxidant effects, but these findings need to be confirmed across a wider range of shrimp species (Kim et al. 2014). Quercetin is the most abundant flavonoid in green tea and appears to have immune-enhancing activity and antioxidant potential in aquatic animals. In shrimp, dietary inclusion of polyphenols such as quercetin can improve growth performance, total haemocyte count (THC) and activities of immune enzymes such as phenoloxidase and LZM. They also have a role in industry stress gene expression, and therefore in the ability for the fish to physically withstand harsh farming conditions. Transcriptomic studies have shown that the shrimp infected by VP had a global downregulation of immune-related genes, especially in susceptible families, indicating an inhibition of vital mechanisms of physiological defence (Liu, Wang, et al. 2023, D. Ma et al. 2020). Moreover, *Vibrio harveyi* infection has been reported to stimulate energy, amino acid and carbohydrate metabolism pathways in shrimp as part of the host response to stress (M.

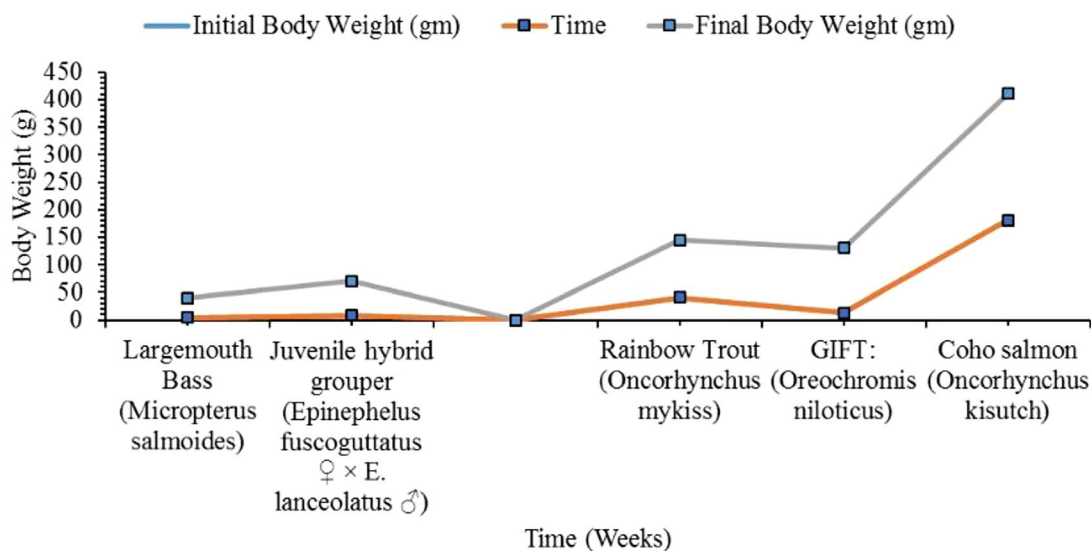


FIGURE 5 | Growth curve of aquaculture species fed green tea—supplemented diets (body weight vs. time). These values were obtained from the following sources (adapted by Yang et al. 2025; Pan et al. 2022; Bilgin, Karabulut, and Tırlı 2025; Daniel et al. 2024; H. Yu et al. 2024).

Sun et al. 2023). Nevertheless, resistant strains, including the Mx family of *L. vannamei*, upregulate transcription of flavonoid biosynthetic genes and have increased tissue levels of quercetin, indicating a role for these bioactive molecules in the modulation of host–pathogen interactions and homeostasis (Aisiah et al. 2022). Some of the promising alternatives used, against increased diseases in intensive shrimp farming (phytochemicals) are tannins, flavonoids and polyphenols in place of the antibiotics (Alfiansah et al. 2020; Okocha et al. 2018; Abidin et al. 2021). Furthermore, the physiological effects of hydrolysable tannins—another group of polyphenols—on shrimp have been shown in various studies. Dietary hydrolysable tannin supplementation has been associated with increased THC, increased LZM activity and the increased expression of antioxidants genes, that is, CAT, GSH-Px and Cu/Zn-SOD, with inclusion levels as low as 0.15% (X. F. Zhu et al. 2021). These responses reflect the enhanced immune defence and alleviation of oxidative stress, which may play important roles in the maintenance of shrimp health in a rearing system with a large population density.

These findings together highlight the role of green TPs, particularly quercetin and tannins, as potential modulators of physiological responses and indicators of improved health and resistance in *Penaeus vannamei*. Their use as feed ingredient in aquafeed formulation appears to be a potential tool for enhancing the shrimp productivity and disease resistance in an ecological and antibiotic-free way.

6 | Conclusion and Future Directions

The integration of *C. sinensis* as a functional feed additive in aquaculture offers significant potential due to its promising immunomodulatory and antioxidant effects, as demonstrated through omics-based approaches. Key bioactive compounds in green tea, particularly catechins, regulate the expression of immune genes (e.g., IL-1 β , TNF- α) and antioxidant genes (e.g., SOD, CAT), thereby enhancing fish health and boosting their

physiological resistance to stress and pathogens. Furthermore, green tea bioactives influence stress-responsive and disease-resistance genes, including HSP70, TLR and NF- κ B, illustrating its positive applications as a natural alternative to antibiotics and synthetic additives. Due to its bioactivity, availability and alignment with sustainable aquaculture practices, *C. sinensis* presents a promising candidate for commercial use. Green tea supplementation provides a sustainable solution for improving growth performance, enhancing disease resistance and promoting the overall well-being of aquaculture species while reducing dependency on antibiotics.

Future research should prioritize dose adjustments, species-specific efficacy and long-term safety evaluations. Further advancements in transcriptomic and metabolomic characterization will aid in deepening the understanding of the molecular mechanisms involved. Large-scale field trials are essential to substantiate the commercial viability of green tea as an aqua-feed additive. In addition, exploring synergistic effects with other natural additives, utilizing highly purified extracts and isolating specific bioactive compounds could lay the groundwork for next-generation functional feeds, tailored to address the evolving challenges faced in aquaculture. Furthermore, long-term ecological assessments are needed to monitor the half-life of green TPs in sediments (estimated to be between 60 and 90 days) and assess the impact of green tea enrichment on benthic communities. Collaborations with ecological economists should be considered to establish an environmental cost-benefit model, quantifying the reduction of carbon footprints such as replacing antibiotics, which could lead to a CO₂ emission reduction of approximately 0.8 tons per hectare of aquaculture water surface.

Author Contributions

Md. Abdullah Al Mamun Hridoy: reviewing and editing the manuscript, conceptualization, methodology, writing – original draft, funding acquisitions. **Azeez Olalekan Baki:** reviewing and editing

manuscript. **Chiara Bordin**: reviewing and editing the manuscript. **Mahedi Hasan**: reviewing and editing the manuscript. **Arman Hos-sain**: reviewing and editing the manuscript. **S. M. Sertaz Islam**: reviewing and editing the manuscript. **Nahida Akter**: writing – original draft. **Md. Masum Reza**: writing – original draft. **Mahima Ranjan Acharjee**: writing – original draft. All authors read the full manuscripts.

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Ethics Statement

This research did not involve human participants, animals, or any direct ethical issues. Therefore, ethical approval was not required for this study.

Consent

The authors have nothing to report.

Conflicts of Interest

The authors declare no conflicts of interest.

Data Availability Statement

No data were used in this research. The study is based on a systematic review and analysis of existing literature and published data.

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