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RESEARCH

Dietary *Saccharomyces cerevisiae* mitigates glyphosate-induced oxidative stress, immunotoxicity and apoptosis in Nile tilapia (*Oreochromis niloticus*)

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Abstract

Saccharomyces cerevisiae (SC) has emerged as a promising probiotic in aquafeeds for enhancing fish growth, health and resilience to environmental stressors. This study investigated the physiological, biochemical, histological and molecular effects of dietary SC supplementation in Nile tilapia (*Oreochromis niloticus*), under normal conditions and following glyphosate (GLY) challenge. Ninety fish (7.93 ± 0.026 g) were randomly allocated into two dietary groups (basal diet and basal diet supplemented with 4 g/kg SC) in triplicate for eight weeks. Following the feeding trial, each group was subdivided into unchallenged and GLY-challenged subgroups (0.6 mg/L; 3.55 µM). SC supplementation significantly improved final body weight, weight gain, specific growth rate and feed conversion ratio, accompanied by upregulation of hepatic insulin-like growth factor 1 (*igf1*) and downregulation of insulin-like growth factor-binding protein 1a (*igfbp1a*) and myostatin (*mstn*). GLY exposure induced hepatic and renal dysfunction, reflected by elevated serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), urea and creatinine, disrupted Lipid and protein profiles, and provoked oxidative stress, inflammation and apoptosis, evident by suppressed nuclear factor erythroid 2–related factor 2 (*nrf2*), superoxide dismutase (*sod*), lysozyme (*lyz*), and complement 3 (*c3*), alongside upregulation of kelch-like ECH-associated protein 1 (*keap1*), tumour necrosis factor-alpha (*tnfα*), cysteine-aspartic acid protease 3 (*cas3*) and cysteine-aspartic acid protease 9 (*cas9*). Histopathological examination confirmed GLY-induced damage in gills, liver and intestinal tissues. Notably, SC supplementation ameliorated these detrimental effects, preserving tissue integrity and restoring molecular and biochemical parameters. These findings highlight the potential of SC as a functional feed additive to enhance performance and mitigate glyphosate-induced toxicity in Nile tilapia, supporting sustainable and resilient aquaculture practices.

Keywords Probiotics · Immune modulation · Aquatic toxicology · Herbicides

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Introduction

Freshwater ecosystems are increasingly threatened by contamination from both anthropogenic and natural sources, posing significant risks to aquatic biodiversity and food security. Such pollutants, including pesticides, heavy metals and other hazardous compounds, can enter aquaculture environments through agricultural runoff, industrial effluents, atmospheric deposition and geological weathering (Mustafa et al. 2024; Lei Zhao et al. 2024; Yu et al. 2021). Among these pollutants, herbicides, most notably glyphosate-based formulations, are recognised as major environmental contaminants due to their widespread use and well-documented toxicity to non-target organisms, including fish (Gandhi et al. 2021; Klátyik et al. 2025; Lopes et al. 2022). Glyphosate [N-(phosphonomethyl)glycine], a broad-spectrum, water-soluble organophosphate herbicide, is widely used for weed control and is among the most frequently detected xenobiotics in surface waters, sediments and effluents (Okayi et al. 2010). Despite its relatively rapid degradation in soil, commercial glyphosate formulations often contain surfactants that enhance herbicidal activity but are highly toxic to aquatic organisms (Tu et al. 2001). Even at sublethal concentrations, glyphosate has been shown to induce a wide range of toxicological effects in fish, including hepatotoxicity, nephrotoxicity, oxidative stress, immune suppression and impaired physiological functions (Helfrich et al. 2009; Hogan 2014). These effects are particularly concerning for aquaculture species, where maintaining water quality, fish health and welfare is essential for optimal productivity and sustainability. The liver and kidney are vital organs in fish, each contributing uniquely to overall physiological function (Mu-Yang Li et al. 2025; Shi et al. 2022). The liver orchestrates metabolic regulation, xenobiotic detoxification, synthesis of biomolecules and immune modulation (Shengqiang Tao et al. 2021; Lei Zhao et al. 2025), while the kidney is primarily responsible for osmoregulation, excretion of metabolic waste and maintaining internal homeostasis (Evans 2010; Lei Zhao et al. 2022). Consequently, glyphosate exposure compromises the integrity and function of these organs, leading to reduced growth, increased disease susceptibility and elevated mortality rates.

Given the increasing prevalence of glyphosate in aquatic environments and its adverse effects on fish health, there is an urgent need for practical, sustainable and cost-effective strategies to mitigate its toxicity. Dietary interventions incorporating functional feed additives such as probiotics, flavonoids, polysaccharides and astaxanthin have emerged as effective strategies to improve fish welfare and enhance resilience to environmental stressors (Abdel-Tawwab and Wafeek 2014; Du et al. 2022; Mu-Yang Li et al. 2024). Probiotics, as defined by the World Health Organisation (WHO) and the Food and Agriculture Organisation (FAO), are "live microorganisms which, when administered in adequate amounts, confer a health benefit on the host" (Hill et al. 2014). In aquaculture, probiotics are widely recognised for their capacity to improve gut health, modulate immune responses, enhance disease resistance and promote physiological homeostasis (Ringø et al. 2022). Among these, *Saccharomyces cerevisiae* (SC), a eukaryotic unicellular yeast, has attracted considerable attention due to its rich nutritional profile, including high-quality protein, essential amino acids, vitamins, enzymes and trace minerals (Solomon et al. 2017). The inclusion of SC and its derivatives in aquafeeds has demonstrated multiple functional benefits, such as immunomodulatory activity, antioxidant capacity and protection against enteric pathogens (Øvrum Hansen et al. 2019; Agboola et al. 2021; Rawling et al. 2019; Vidakovic et al. 2020). These effects are attributed to various mechanisms, including the degradation of toxins, competitive exclusion of pathogens and the enhancement of mucosal barrier integrity through the action of cell wall components like β-glucans and mannan oligosaccharides

(Pérez-Sotelo et al. 2005; Fakruddin et al. 2017; Arévalo-Villena et al. 2018; Roussel et al. 2018; Štofilová et al. 2022).

Nile tilapia (*Oreochromis niloticus*) is one of the most widely farmed fish species worldwide, valued for its adaptability, rapid growth and disease resistance, contributing to a global market estimated at approximately USD 13.8 billion (El-Sayed 2020; FAO 2025). Egypt is among the world's top producers of Nile tilapia, playing a crucial role in both national food security and global aquaculture supply. The country's tilapia production has reached approximately 1.1 million metric tons annually (FAO 2025). However, a substantial portion of Egyptian aquaculture depends on agricultural drainage water, which increases the risk of contamination with agrochemical residues, including glyphosate (Ghanem and Haggag 2015). A recent field survey in Ismailia Governorate detected glyphosate residues in water samples from four out of five tilapia farms, with concentrations ranging from 0.813 to 5.431 µg/L, indicating a potential ecological threat to aquatic organisms (Hassan et al. 2022). While several studies have reported the growth-promoting and immunostimulatory effects of dietary yeast supplementation in fish (Abdel-Tawwab et al. 2020; Xuanyi Yang et al. 2020; Agboola et al. 2021), the mechanisms by which it may counteract glyphosate-induced toxicity in tilapia remain poorly understood. Therefore, the present study aims to investigate the potential protective effects of dietary *Saccharomyces cerevisiae* supplementation in Nile tilapia exposed to glyphosate, focusing on its influence on oxidative stress, hepatic and renal dysfunction, inflammation, apoptosis and the expression of tight junction-related genes.

Materials and methods

Ethical statement

All animal procedures in this study were carried out in accordance with the Egyptian Code of Ethics for the Use of Animals in Research. The experimental protocol was reviewed and approved by the Animal Ethics Committee of the Faculty of Aquatic and Fisheries Sciences, Kafrelsheikh University, Egypt (Approval No: IAACUC-KSU-3–2022).

Feed additive and experimental diets preparation

The feed additive used in this study was a commercial formulation of *Saccharomyces cerevisiae* (SC), containing viable yeast cells standardised for use in aquafeeds. It was incorporated into the basal diet at a concentration of 4 g/kg feed, based on previous recommendations for functional supplementation in fish. According to the manufacturer's specifications, this commercial formulation primarily contains viable *S. cerevisiae* cells, with trace amounts of emulsifier and ascorbic acid. The basal diet was formulated to meet the nutritional requirements of Nile tilapia, containing 31.8% crude protein and an estimated digestible energy of approximately 3005 kcal/kg, following NRC guidelines. All ingredients were finely ground and thoroughly mixed. Two experimental diets were prepared: one control diet without supplementation and one supplemented with 4 g/kg *S. cerevisiae* (Table 1). The mixed diets were pelletised using a meat grinder fitted with a 2.33 mm die, shade-dried and stored at 4 °C until use to prevent nutrient degradation and oxidative rancidity.

Table 1 Ingredients and proximate composition (% as-fed basis) of the basal and *Saccharomyces cerevisiae*-supplemented (SC) diets

Ingredients (% as feed)	Basal	SC
Fish meal (65%)	10	10
Soybean meal (45%)	40.8	40.8
Corn gluten meal	6	6
Yellow corn	19.5	20.2
Wheat flour	18.5	16.8
Soybean oil	3	3
Vitamin premix *	0.8	0.8
Mineral premix **	0.5	0.5
DiCaP	0.6	0.6
Choline chloride	0.2	0.2
Stay C®***	0.1	0.1
Saccharomyces****	0	4
Composition (%)		
Crude protein	31.85	31.71
DE (Kcal/Kg)	3005.16	2999.58
Crude lipid	5.44	5.44
Ash	4.923	4.926
Crude fiber	3.827	3.821
Ca	0.784	0.784
P	0.810	0.806

*Vitamin premix (g/kg): Thiamin HCl (0.44), Riboflavin (0.63), Pyridoxine HCl (0.91), DL pantothenic acid (1.72), Nicotinic acid (4.58), Biotin (0.21), Folic acid (0.55), Inositol (21.05), Menadione sodium bisulfite (0.89), Vitamin A acetate (0.68), Vitamin D3 (0.12), dL-alpha-tocopherol acetate (12.63), Alpha-cellulose (955.59)

**Mineral premix (g/100 g): Cobalt chloride (0.004), Cupric sulfate pentahydrate (0.25), Ferrous sulfate (4.00), Magnesium sulfate anhydrous (13.862), Manganous sulfate monohydrate (0.650), Potassium iodide (0.067), Sodium selenite (0.010), Zinc sulfate heptahydrate (13.193), Alpha-cellulose (67.964)

***Stay C®, (L-ascorbyl-2-polyphosphate 35%)

**** *Saccharomyces cerevisiae* 4 g/kg

Experimental design and rearing conditions

A total of 90 apparent 'healthy' mono-sex male Nile tilapia (*Oreochromis niloticus*) with an initial average body weight of 7.93 ± 0.026 g were obtained from a local private fish farm in Kafrelsheikh Governorate, Egypt. Fish were transported in oxygenated polyethylene bags to the Biotechnology Laboratory Aquarium Facility, Faculty of Aquatic and Fisheries Sciences, Kafrelsheikh University. Upon arrival, fish were acclimated for two weeks and fed a standard commercial diet at 5% of their body weight, administered twice daily.

Following acclimation, fish were randomly assigned to two dietary treatment groups with three replicates per group (15 fish per $70 \times 60 \times 40$ cm glass tank). The feeding trial continued for 8 weeks. Tanks were equipped with continuous aeration (air stones) and mechanical filtration. Water quality parameters were monitored daily using calibrated portable instruments. Total ammonia nitrogen (TAN) was measured weekly using a HI-97715 photometer® (Hanna Instruments Ltd., UK), employing the standardised

Nessler method. DO was assessed using a HI-9146–04 HANNA® galvanic DO meter with automatic temperature compensation, and pH/temperature were recorded using an MW105 portable meter® (Milwaukee Instruments, Hungary). The recorded values were: temperature 26.23 ± 3 °C, dissolved oxygen (DO) 5.8 ± 0.7 mg/L and pH 7.60 ± 0.2.

Growth performance evaluation

Fish were weighed at the beginning of the experiment and biweekly afterwards. Growth performance indices were calculated at the end of the feeding trial as follows:

- Weight Gain (WG) = Final BW − Initial BW
- Percent Weight Gain (PWG) = [(Final BW − Initial BW)/Initial BW] × 100
- Specific Growth Rate (SGR) = [(ln Final BW − ln Initial BW)/Days] × 100
- Feed Conversion Ratio (FCR) = Feed Intake (g)/Weight Gain (g)
- Protein Efficiency Ratio (PER) = Weight Gain (g)/Protein Intake (g)
- Survival Rate (%) = (Final number of fish/Initial number of fish) × 100

Glyphosate challenge

Following the 8-week feeding trial, each dietary group was subdivided into challenged and non-challenged subgroups: CON (control, non-challenged), CON + GLY (control, glyphosate-challenged), SC (*Saccharomyces cerevisiae*-supplemented, non-challenged), and SC + GLY (*Saccharomyces cerevisiae*-supplemented, glyphosate-challenged). Challenged groups were exposed to a commercial glyphosate formulation (Roundup® 48%, Agrochem, Alwatneia Co., Alexandria, Egypt) at a concentration of 0.6 mg/L (3.55 µM). This sublethal concentration (1/20th of the LC50, 12 mg/L) was based on previous findings (Abdelmagid et al. 2022, 2021) to induce measurable physiological and biochemical responses without inducing acute mortality. Glyphosate was administered via waterborne exposure for three consecutive days, with daily renewal of treated water. Control tanks received dechlorinated tap water.

Blood and tissue sampling

Following the challenge experiment, 5 fish from each tank (15 per group) were anaesthetised with MS-222 (150 mg/L; Argent Laboratories, Redmond, WA, USA). Blood was collected from the caudal vein into microcentrifuge tubes without anticoagulant, allowed to coagulate, and centrifuged at 3000 rpm for 15 min at 4°C. Serum samples were aliquoted and stored at − 20 °C until biochemical analyses. Tissue samples (gills, liver and intestine) were excised immediately post-blood collection. Samples were fixed in 10% neutral buffered formalin for histopathology, whereas gene expression samples were snap-frozen in liquid nitrogen and stored at − 80°C.

Serum biochemical analysis

Serum parameters were assessed using spectrophotometry and commercial kits (Bio-Diagnostic, Cairo, Egypt). Total protein and albumin levels were determined following Lowry et al. (1951) and Doumas et al. (1971), respectively. Total cholesterol (TC) and triglycerides (TG) were measured per Allain et al. (1974) and Bucolo and David (1973). Blood urea nitrogen (BUN) and creatinine were determined based on (Patton and Crouch 1977) and Henry (1974). Alanine Transaminase (ALT) and Aspartate Aminotransferase (AST) levels were analysed according to Reitman and Frankel (1957). Very low-density lipoprotein (VLDL-C) was calculated using the Friedewald equation (Friedewald et al. 1972). Globulin was calculated by subtracting albumin from total protein, and the albumin/globulin ratio (A/G) was determined per Kaneko (1989).

Molecular analysis

Total RNA was extracted from liver and intestine samples (n = 15 per group; 50 mg tissue each) using GENEzol™ Reagent (Geneaid, UK), following the manufacturer's protocol. RNA concentration and purity were assessed using a BioDrop spectrophotometer (Biochrom Ltd., UK), and RNA integrity was verified by agarose gel electrophoresis. Five micrograms of total RNA were reverse-transcribed into cDNA using the TOPscript™ RT DryMIX kit (Enzynomics, Korea). Quantitative real-time PCR (qPCR) was performed using TOPreal™ SYBR Green qPCR PreMIX (Enzynomics, Korea) and gene-specific primers targeting genes associated with growth, antioxidant, inflammation, apoptosis and tight junction integrity. The genes analysed included insulin-like growth factor 1 (*igf1*), insulin-like growth factor binding protein 1a (*igfbp1a*), myostatin (*mstn*), nuclear factor erythroid 2–related factor 2 (*nrf2*), kelch-like ECH-associated protein 1 (*keap1*), superoxide dismutase (*sod*), tumour necrosis factor-alpha (*tnfα*), complement 3 (*c3*), lysozyme (*lyz*), cysteine-aspartic acid protease 3 (*cas3*), cysteine-aspartic acid protease 9 (*cas9*), occludin (*ocln*) and claudin3 (*cldn3*) (Table 2). A 20 µL qPCR reaction consisting of 0.6 µL forward primer, 0.6 µL reverse primer, 1 µL cDNA, 7.8 µL nuclease-free water and 10 µL SYBR Green master mix was prepared in triplicate for each sample. qPCR was carried out on a Rotor-Gene Q cycler (QIAGEN, Germany) under the following cycling conditions: initial activation at 95 °C for 15 min, followed by 40 cycles of denaturation at 95 °C for 10 s, annealing at gene-specific temperatures for 15 s, and extension at 72 °C for 25 s. Melt curve analysis was conducted to verify amplification specificity. Relative gene expression levels were calculated using the $2^{-\Delta\Delta CT}$ method (Livak and Schmittgen 2001), normalised against housekeeping genes elongation factor 1 alpha (*ef1a*) and glyceraldehyde-3-phosphate dehydrogenase (*gapdh*).

Histopathological examination

Fifteen fish per group were examined histologically. Gills, Liver and intestine were fixed in 10% neutral buffered formalin (Thermo Fisher, USA) for 48 h. Tissues were rinsed, dehydrated in 75% ethanol, embedded in paraffin, sectioned at 5 µm, fixed on glass slides and stained with hematoxylin and eosin (H&E) according to Gamble (2008) Microscopic examination was performed under a standard light microscope.

Table 2 Primer sequences used for quantitative real-time PCR (qPCR)

	Gene	Primer sequence (5'–3')	Accession number	Annealing temperature (°C)	Reference
1	*ef1a*	F: GCACGCTCTGCTGGCCTTT R: AGCCAGACGGACAGATGCC	AB075952	59	Chang Geng Yang et al. (2013)
2	*gapdh*	F: GATAATGGCAAACTTGTCGTCG R: ACATTGGAGCATCGGGTGAG	JN381952		
3	*igf1*	F: TCCTGTAGCCACACCCTCTC R: ACAGCTTTGGAAGCAGCACT	NM_001279503.1	56	Costa et al. (2016)
4	*igfbp1a*	F: TCCTAGACCTGGTGAAGCCA R: CGAGGTCG ACAGTGC AGATT	XM_003438121.3		Assar et al. (2025)
5	*mstn*	F: GCATCTGTCTCAGATCGTGCT R: TGCCATCATTACAATTGTCTCCG	KT987208.1	60	Elkatatny et al. (2016)
6	*nrf2*	F: CTGCCGTAAACGCAAGATGG R: ATCCGTTGACTGCTGAAGGG	XM_003447296.4	57	Elbialy et al. (2024)
7	*keap1*	F: CTTCGCCATCATGAACGAGC R: CACCAACTCCATACCGCACT	XM_003447926.3		
8	*sod*	F: CATGCCTTCGGAGACAACAC R: ACCTTCTCGTGGATCACCAT	XM_003446807.5	57	Rashwan et al. (2024)
9	*c3*	F: GGTGTGGATGCACCTGAGAA R: GGGAAATCGGTACTTGGCCT	XM_013274267.2	57	Jia et al. (2019b)
10	*lyz*	F: AAGGGAAGCAGCAGCAGTTGTG R: GTCCATGCCGTTAGCCTTGAG	XM_003460550.2	61	
11	*tnfα*	F: GGAAGCAGCTCCACTCTGATGA R: CACAGCGTGTCTCCTTCGTTCA	NM_001279533.1	61	Rashwan et al. (2024)
12	*cas3*	For: GGCTCTTCGTCTGCTTCTGT Rev: GGGAAATCGAGGCGGTATCT	GQ421464.1	60	Standen et al. (2016)
13	*cas9*	F: CTTCAGCGGAACAGGGTTA R: GAAGGCACTCCAGAAATAAGG	XM_025901776.1		Y Zheng et al. (2016)

Table 2 (continued)

	Gene	Primer sequence (5'–3')	Accession number	Annealing temperature (°C)	Reference
14	*ocln*	AATCGGGATAATCTCCTACA TTGGTCCTCTTTGCTATTTG	XM_003438009.5	57	Hongqin Li et al. (2019)
15	*cldn3*	F: GCAACATTGTGACGGCTCAGAT R:AGAGGGCGAGCATAGAGTCATACA	XM_005465025.3		Yu-Xue Zhang et al. (2020)

ef1a: elongation factor 1 alpha (housekeeping gene), ***gapdh***: glyceraldehyde-3-phosphate dehydrogenase (housekeeping gene), ***igf1***: insulin-like growth factor 1, ***igfbp1a***: insulin-like growth factor binding protein 1a, ***mstn***: myostatin, ***c3*:** complement 3, ***lzy***: lysozyme, ***sod***: superoxide dismutase, ***nrf2***: nuclear factor erythroid 2–related factor 2, ***keap1***: kelch-like ECH-associated protein 1, ***tnfα***: tumer necrosis factor alpha, ***cas3***: cysteine-aspartic acid protease 3, ***cas* 9**: cysteine-aspartic acid protease 9, ***ocln*:**: occludin, ***cldn3***: claudin3.

Statistical analysis

All data were expressed as mean ± standard error of the mean (SEM). Data normality was assessed using Shapiro–Wilk's test and homogeneity of variances was tested with Levene's test. Arcsine transformation was applied to percentage data before analysis. One-way ANOVA was used to evaluate growth performance, and two-way ANOVA followed by Tukey's post hoc test was used for biochemical, histopathological and gene expression data ($p < 0.05$). Statistical analyses were conducted using GraphPad Prism version 9.5 (GraphPad Software, San Diego, CA, USA).

Results

Growth indices and molecular markers of growth

Growth performance of Nile tilapia was significantly influenced by dietary *Saccharomyces cerevisiae* (SC) supplementation ($p < 0.05$; Table 3). Fish fed the SC-supplemented diet exhibited a significantly higher final body weight (FBW), body weight gain percentage (BWG%) and specific growth rate (SGR) compared to the control group ($p = 0.04$, 0.045 and 0.02, respectively). Feed efficiency was also significantly improved, as reflected by a lower feed conversion ratio (FCR) and higher protein efficiency ratio (PER) in the SC group compared to control ($p = 0.01$ and 0.03, respectively). No significant differences were observed in survival rate or total feed intake between groups ($p > 0.05$).

At the molecular level, the hepatic expression of growth-related genes further supported the enhanced growth performance (Fig. 1). SC supplementation significantly upregulated *igf1* expression in both SC and SC + GLY groups ($p < 0.05$). Conversely, *igf1* expression was markedly downregulated in fish exposed to glyphosate (CON + GLY), confirming its growth-suppressive effect. Additionally, *igfbp1a* and *mstn* expression levels were significantly downregulated in the SC group compared to CON, while CON + GLY fish showed upregulated expression of these genes. These results suggest that SC enhances growth in

Table 3 Growth performance and feed utilisation parameters of Nile tilapia (*Oreochromis niloticus*) fed control or *Saccharomyces cerevisiae*-supplemented diets over an 8-week period

Parameters	Control	SC	*p* value
IBW (g)	7.91 ± 0.002	7.96 ± 0.003	0.52
FBW (g)	24.42 ± 0.02[b]	26.07 ± 0.01[a]	0.04
Total feed intake (g)	375.76 ± 2.10	375.76 ± 2.10	> 0.99
BWG %	208.75 ± 6.75[b]	229.47 ±2.61[a]	0.045
SGR (% day^{-1})	1.87 ± 0.03 [b]	2 .00± 0.00[a]	0.02
FCR	1.82 ± 0.02 [a]	1.59 ± 0.04[b]	0.01
PER	1.77 ± 0.03 [b]	2.00 ± 0.06[a]	0.03
Survival (%)	88.9 ±2.2	91.1 ±2.2	0.52

IBW: Initial body weight; *FBW*: Final body weight; *BWG*: Body weight gain; *SGR*: Specific growth rate; *FCR*: Feed conversion ratio; *PER*: Protein efficiency ratio. Data are expressed as Mean ± SEM where n = 3 as triplicate tanks for BWG%, FCR, SGR, PER and n = 45 for IBW and FBW. Different superscript letters within the same row indicate statistically significant differences ($p < 0.05$).

Fig. 1 Relative expression of growth-related genes in the liver of Nile tilapia (*Oreochromis niloticus*) fed a control or *Saccharomyces cerevisiae*-supplemented diet, under non-challenged and glyphosate-challenged conditions. **A** *igf1* (insulin-like growth factor 1), **B** *igfbp1a* (insulin-like growth factor binding protein 1a) and (**C**) *mstn* (myostatin). Data are presented as mean ± SEM (n = 15/group). Columns with different superscript letters indicate statistically significant differences between groups ($p < 0.05$)

Nile tilapia by modulating endocrine growth pathways and repressing growth-inhibitory genes.

Biochemical findings

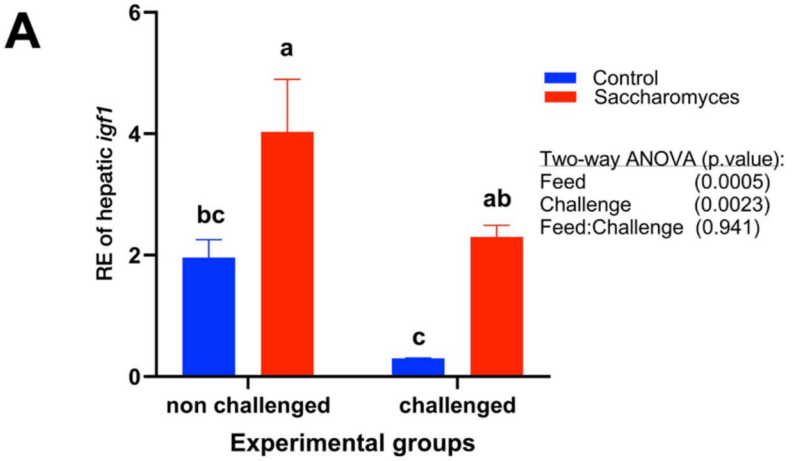
Serum total protein and globulin levels were significantly increased in both SC-supplemented groups compared to the CON and CON + GLY groups ($p < 0.0001$), as shown in Table 4. Albumin levels also followed this trend and were significantly higher in the SC-fed groups ($p = 0.0003$), while the albumin/globulin (A/G) ratio was slightly but significantly reduced in the SC-fed groups ($p = 0.0120$) (Table 4). In the lipid profile, glyphosate exposure resulted in a marked elevation in serum cholesterol, triglycerides and very-low-density lipoprotein (VLDL) levels in the CON + GLY group ($p < 0.0001$; Table 4). In contrast, SC-fed fish exhibited significantly lower lipid levels compared to CON. Notably, fish in the SC + GLY group showed partially restored lipid profiles relative to the CON + GLY group, highlighting the ability of SC to mitigate glyphosate-induced lipid imbalances. Renal function markers, including creatinine and blood urea nitrogen (BUN), were markedly elevated in the CON + GLY group, indicating the nephrotoxic effects of glyphosate ($p < 0.0001$; Table 4). In contrast, SC supplementation significantly reduced creatinine and BUN concentrations in the SC + GLY group compared to the CON + GLY group, indicating a nephroprotective effect of SC. Notably, the SC group maintained normal renal marker levels, comparable to the control. Similarly, hepatic enzyme activities (ALT and AST) were markedly increased in the CON + GLY group ($p < 0.0001$; Table 4), confirming glyphosate-induced hepatic injury. In the SC + GLY group, these enzyme levels were significantly lower than those in the CON + GLY group, suggesting a hepatoprotective effect of SC. Importantly, SC supplementation alone did not induce any hepatic stress, as enzyme levels remained comparable to the control group.

Differential gene expression analysis

Dietary supplementation with SC significantly modulated the hepatic and intestinal expression of genes related to oxidative stress response, inflammation, immune function, apoptosis and intestinal barrier integrity in Nile tilapia. The effects were evident across both non-challenged and glyphosate (GLY)-challenged conditions, as detailed below.

Hepatic antioxidant gene expression

SC supplementation significantly upregulated the hepatic expression of *nrf2* and *sod* in both SC and SC + GLY groups compared to the CON and CON + GLY groups ($p < 0.05$; Fig. 2D, 2E). GLY exposure (CON + GLY) markedly suppressed *nrf2* and *sod* expression, reflecting oxidative stress induction. *Keap1*, a negative regulator of *nrf2*, was significantly upregulated in the CON + GLY group and downregulated in SC-fed fish ($p < 0.05$: Fig. 2F),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ab
non challenged
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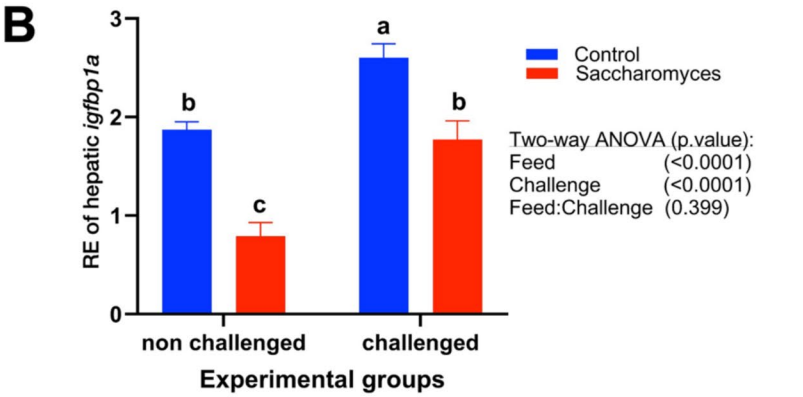
Experimental groups
Control
Saccharomyces
Two-way ANOVA (p.value):
Feed (0.0005)
Challenge (0.0023)
Feed:Challenge (0.941)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RE of hepatic igfbp1a
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non challenged
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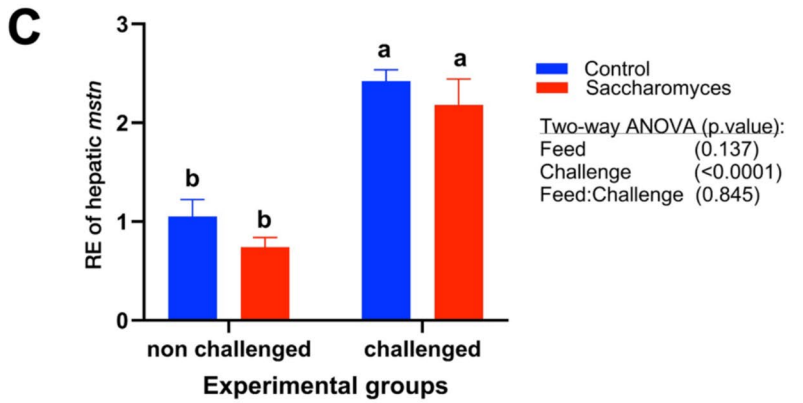
Experimental groups
Control
Saccharomyces
Two-way ANOVA (p.value):
Feed (<0.0001)
Challenge (<0.0001)
Feed:Challenge (0.399)


C
RE of hepatic mstn
3
2
1
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b
non challenged
challenged
Experimental groups
Control
Saccharomyces
Two-way ANOVA (p.value):
Feed (0.137)
Challenge (<0.0001)
Feed:Challenge (0.845)

Table 4 Biochemical parameters of Nile tilapia (*Oreochromis niloticus*) fed a control or *Saccharomyces cerevisiae*-supplemented diet over an 8-week period, with or without glyphosate challenge. Parameters assessed include total protein, cholesterol, triglycerides, very low-density lipoprotein (VLDL), urea, creatinine, glucose, albumin, globulin, albumin/globulin (A/G) ratio, aspartate aminotransferase (AST) and alanine aminotransferase (ALT). Data are expressed as mean ± SEM (n = 15/group). Different superscript letters within rows indicate significant differences (two-way ANOVA, $p < 0.05$). All parameters were measured in serum

Parameters	Control	Saccharomyces	Control challenged	Saccharomyces challenged	*p* value of two-way ANOVA		
					Challenge	Treatment	Interaction
Total protein (g/dL)	4.62 ± 0.011^{b}	4.91 ± 0.023^{a}	4.566 ± 0.0098^{b}	4.86 ± 0.024^{a}	0.0596	< 0.0001	0.8365
Albumin (g/dL)	2.52 ± 0.005^{bc}	2.66 ± 0.044^{a}	2.5 ± 0.004^{c}	2.62 ± 0.034^{ab}	0.3160	0.0003	0.8210
Globulins (g/dL)	1.95 ± 0.95^{b}	2.25 ± 0.067^{a}	1.934 ± 0.001^{b}	2.16 ± 0.029^{b}	0.1863	< 0.0001	0.3523
A/G	1.294 ± 0.02	1.19 ± 0.055	1.292 ± 0.00198	1.22 ± 0.03	0.6995	0.0120	0.6636
VLDL (mg/dL)	22.689 ± 0.088^{b}	18.6 ± 0.057^{c}	24.88 ± 1.88^{a}	23.15 ± 0.11^{b}	< 0.0001	< 0.0001	< 0.0001
Cholesterol (mg/dL)	105.04 ± 0.050^{c}	99.85 ± 0.01^{d}	115.87 ± 0.497^{a}	109.28 ± 0.33^{b}	< 0.0001	< 0.0001	0.5121
Triglycerides (mg/dL)	113.44 ± 0.44^{b}	93.03 ± 0.28^{c}	124.4 ± 0.41^{a}	115.77 ± 0.55^{b}	< 0.0001	< 0.0001	< 0.0001
Creatinine (mg/dL)	0.87 ± 0.002^{c}	0.75 ± 0.335^{d}	1.13 ± 0.003^{a}	0.99 ± 0.012^{b}	< 0.0001	< 0.0001	0.2407
Blood Urea nitrogen (mg/dL)	15.136 ± 0.005^{c}	13.28 ± 0.246^{d}	19.203 ± 0.009^{a}	16.26 ± 0.125^{b}	< 0.0001	< 0.0001	0.0014
Ast (U/L)	35.36 ± 0.214^{c}	32.45 ± 0.178^{d}	56.6 ± 0.21^{a}	39.15 ± 0.42^{b}	< 0.0001	< 0.0001	< 0.0001
Alt (U/L)	28.193 ± 0.011^{c}	25.55 ± 0.48^{d}	43.266 ± 0.1747^{a}	35.31 ± 0.45^{b}	< 0.0001	< 0.0001	< 0.0001

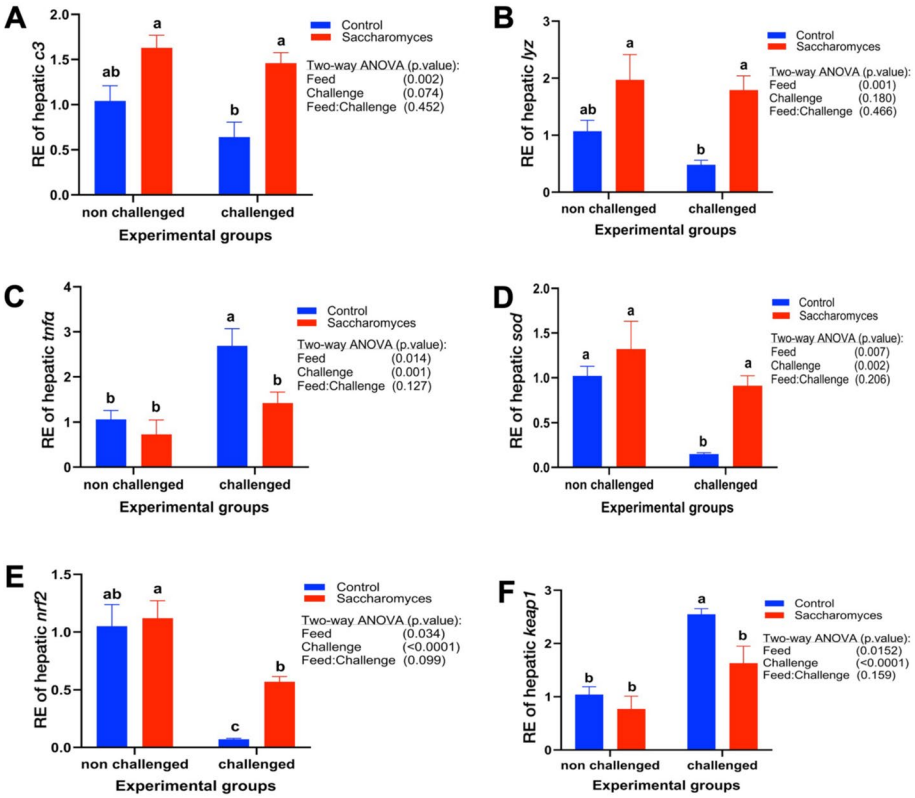

Fig. 2 Relative expression of immune and antioxidant-related genes in the liver of Nile tilapia (*Oreochromis niloticus*) fed a control or *Saccharomyces cerevisiae*-supplemented diet, under non-challenged and glyphosate-challenged conditions. **A** *c3* (complement 3), **B** *lyz* (lysozyme), **C** *tnfα* (tumor necrosis factor-alpha), **D** *sod* (superoxide dismutase), **E** *nrf2* (nuclear factor erythroid 2-related factor 2) and (**F**) *keap1* (kelch-like ECH-associated protein 1). Data are presented as mean ± SEM (n = 15/group). Columns with different superscript letters indicate statistically significant differences between groups ($p < 0.05$)

suggesting that SC counteracts GLY-induced oxidative imbalance by promoting *nrf2*-mediated signalling.

Hepatic inflammatory and immune gene expression

GLY exposure (CON + GLY) significantly upregulated the expression of *tnfα* (Fig. 2C), indicating a pro-inflammatory effect of glyphosate. In contrast, SC-fed groups (SC and SC + GLY) exhibited significantly lower hepatic *tnfα* expression levels ($p < 0.05$). Additionally, the immune-related genes *lyz* and *c3* were significantly upregulated in SC and SC + GLY groups ($p < 0.05$; Figs. 2A-B), highlighting the immunostimulatory effect of SC. These genes were downregulated in the CON + GLY group, suggesting GLY-induced suppression.

Intestinal apoptosis-related gene expression

Markers of apoptosis, including *casp3* and *casp9*, were significantly elevated in the CON + GLY group ($p < 0.05$), reflecting increased intestinal cell apoptosis under glyphosate stress (Fig. 3A-B). However, SC supplementation significantly reduced *cas3* and *cas9* expression in both SC and SC + GLY groups ($p < 0.05$), indicating an anti-apoptotic effect. Notably, the SC + GLY group exhibited intermediate expression levels between SC and CON + GLY, suggesting that SC potentially mitigates glyphosate-induced apoptosis.

Intestinal tight junction gene expression

Expression of intestinal barrier genes *ocln* and *cldn3* was significantly reduced in the CON + GLY group ($p < 0.05$), indicating impaired intestinal integrity (Fig. 3C–D). While there were no significant differences between SC and CON groups, SC + GLY fish displayed significantly higher *ocln* and *cldn3* expression compared to CON + GLY ($p < 0.05$), suggesting a protective effect of SC against GLY-induced barrier dysfunction.

Histopathological findings

Histopathological examination of gill, liver and intestinal tissues revealed marked structural alterations in response to glyphosate (GLY) exposure and potential protective effects of *Saccharomyces cerevisiae* (SC) supplementation, as illustrated in Figs. 4–6. Gill sections from the CON group displayed normal histological features, including well-defined

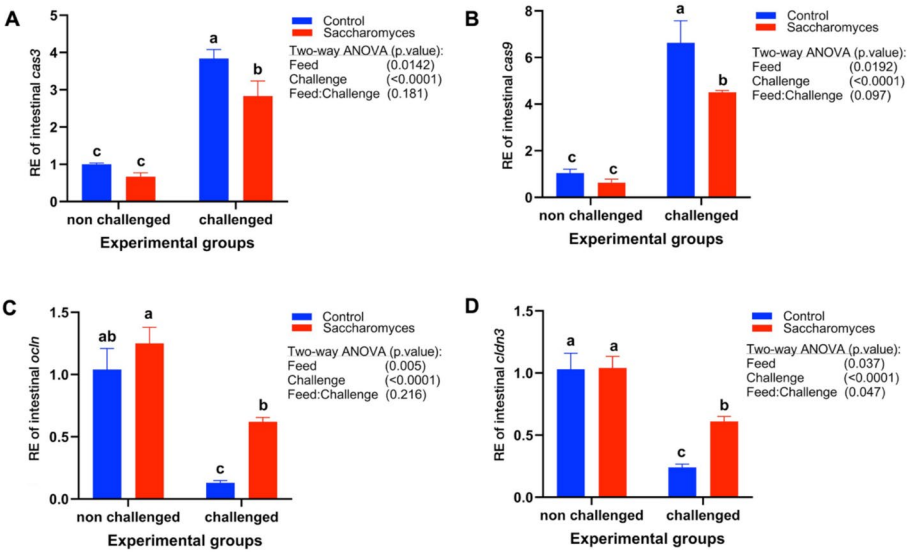

Fig. 3 Relative expression of apoptosis-related and tight junction-related genes in the intestine of Nile tilapia (*Oreochromis niloticus*) fed a control or *Saccharomyces cerevisiae*-supplemented diet, under challenged and non-challenged conditions. **A** *cas3* (cysteine-aspartic acid protease 3), **B** *cas9* (cysteine-aspartic acid protease 9), **C** *ocln* (occludin) and (**D**) *cldn3* (claudin3). Data are presented as mean ± SEM (n = 15/group). Columns with different superscript letters indicate significant differences ($p < 0.05$)

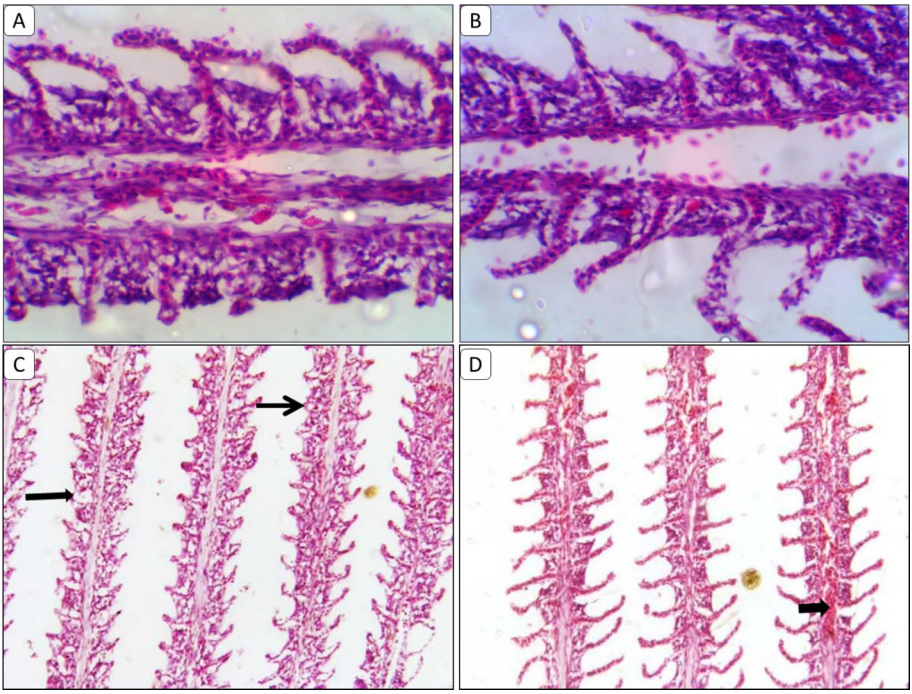

Fig. 4 Representative photomicrographs of gill tissues from Nile tilapia fed a control or *Saccharomyces cerevisiae*-supplemented diet, under challenged and non-challenged conditions. **A** Control group (CON) showing normal histological architecture of primary and secondary lamellae. **B** Glyphosate-challenged group (CON + GLY) exhibiting diffuse lamellar fusion (thin arrow), congestion and hemorrhage (thick arrow). **C** *Saccharomyces cerevisiae*-supplemented group (SC) showing mild to moderate lamellar fusion (thin arrow) and lamellar lifting (thick arrow). **D** *Saccharomyces cerevisiae*-supplemented challenged group (SC + GLY) displaying minimal congestion (thick arrow) with normal intact lamellar structure

primary and secondary lamellae without any pathological lesions (Fig. 4A). In contrast, GLY exposure (CON + GLY) induced prominent lesions such as interlamellar epithelial hyperplasia, fusion of secondary lamellae, mucous cell proliferation, detachment and aneurysm of the lamellar epithelium (Fig. 4B). SC-supplemented fish (SC) retained a normal gill architecture, indicating no adverse effects of SC on gill morphology (Fig. 4C). Notably, SC + GLY fish exhibited substantially reduced lamellar hyperplasia, mild mucous cell proliferation and limited epithelial damage, with better lamellar integrity compared to CON + GLY, suggesting a protective effect of SC against GLY-induced gill injury (Fig. 4D). Liver sections from the CON group demonstrated healthy hepatic parenchyma with intact hepatocytes and pancreatic acini (Fig. 5A). In contrast, livers from CON + GLY fish showed severe hepatic damage, including diffuse coagulative necrosis, severe fatty vacuolation, inflammatory cell infiltration, sinusoidal congestion and pyknotic nuclei. Additionally, the pancreatic acinar cells exhibited cytoplasmic degeneration and loss of zymogen granules (Fig. 5B-C). SC-fed fish (SC group) exhibited near-normal hepatic architecture with mild cytoplasmic vacuolation and no evidence of inflammation or necrosis (Fig. 5D). Importantly, SC + GLY fish presented with markedly reduced hepatocyte vacuolisation and sinusoidal congestion, with better-preserved pancreatic acinar structures and reduced inflammatory infiltration compared to the CON + GLY group (Fig. 5E),

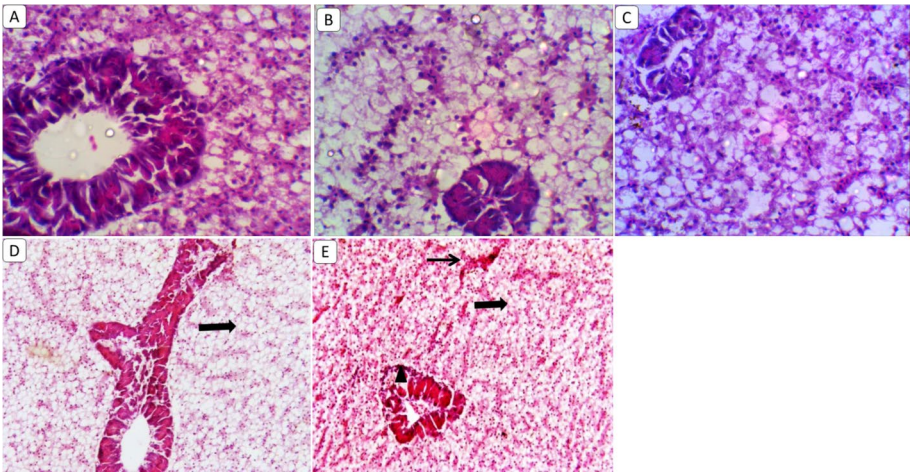

Fig. 5 Representative photomicrographs of liver tissues from Nile tilapia fed a control or *Saccharomyces cerevisiae*-supplemented diet, under challenged and non-challenged conditions. **A** Control group (CON) showing normal histological architecture of hepatic parenchyma and hepatopancreas. **B**-**C** Glyphosate-challenged group (CON + GLY) displaying swollen hepatocytes (thick arrow), multifocal coalescing hepatocellular necrosis (arrowhead) and inflammatory cell aggregates (thin arrow). **D** *Saccharomyces cerevisiae*-supplemented group (SC) exhibiting moderate to severe hepatic vacuolation (thick arrow). **E** *Saccharomyces cerevisiae*-supplemented challenged group (SC + GLY) showing mild hepatic vacuolation (thick arrow), sinusoidal congestion (thin arrow) and minimal perihepatopancreatic inflammatory aggregates (arrowhead)

indicating the hepatoprotective role of SC under GLY challenge. Intestinal sections from the CON group revealed well-organised villi with intact epithelial linings and balanced goblet cell distribution (Fig. 6A). GLY exposure (CON + GLY) caused extensive mucosal damage, including severe intestinal epithelial desquamation, villus blunting, crypt shortening, submucosal oedema, infiltration of the lamina propria with inflammatory cells, and in some cases, congestion and haemorrhage (Fig. 6B-C). The SC group exhibited preserved intestinal morphology, with intact villi and minimal histopathological alterations (Fig. 6D). The SC + GLY group showed significant structural recovery, demonstrating only mild submucosal oedema and reduced inflammatory infiltration and necrotic changes compared to the CON + GLY group (Fig. 6E), further confirming the beneficial effects of SC supplementation in maintaining intestinal integrity under GLY-induced stress.

Discussion

The current study demonstrated that dietary supplementation with *Saccharomyces cerevisiae* (SC) significantly enhanced growth performance, immune function, oxidative status and histopathological integrity in Nile tilapia, particularly under glyphosate (GLY) exposure. These findings highlight the potential of SC as a functional feed additive in mitigating herbicide-induced toxicity in aquaculture.

SC-supplemented diets significantly improved final body weight, weight gain percentage, specific growth rate (SGR), feed conversion ratio (FCR) and protein efficiency ratio (PER), indicating enhanced growth performance. These effects can be attributed to the presence of bioactive components in yeast such as β-glucans, mannan oligosaccharides

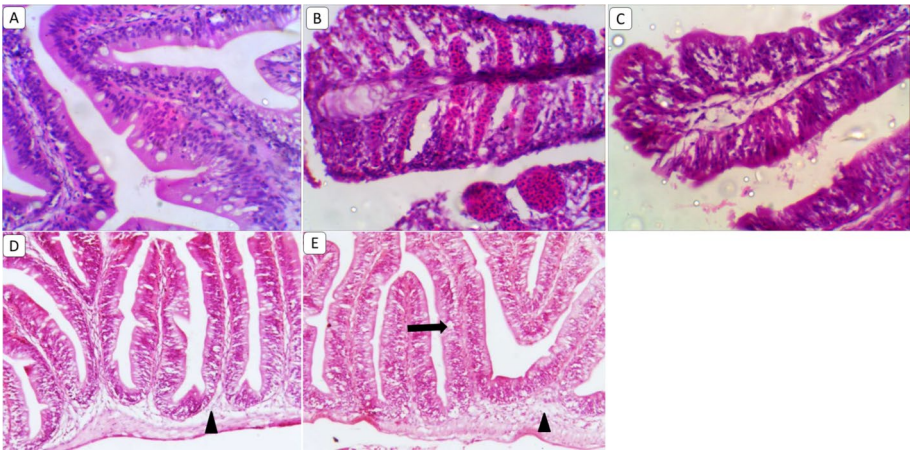

Fig. 6 Representative photomicrographs of intestinal sections from Nile tilapia fed a control or *Saccharomyces cerevisiae*-supplemented diet, under challenged and non-challenged conditions. **A** Control group (CON) displaying normal histological architecture of intestinal villi. **B**-**C** Glyphosate-challenged group (CON + GLY) showing extensive apical desquamation (thin arrow) and mild lamina propria inflammatory aggregates (arrowhead). **D** *Saccharomyces cerevisiae*-supplemented group (SC) with minimal to mild submucosal edema (arrowhead). **E** *Saccharomyces cerevisiae*-supplemented challenged group (SC + GLY) exhibiting mild mucosal vacuolation (thick arrow) and mild submucosal edema (arrowhead)

(MOS), nucleotides and essential nutrients, which are known to promote digestive enzyme activity, nutrient absorption and gut microbial balance (Huyben et al. 2017; Adel et al. 2017; Øvrum Hansen et al. 2019; Vidakovic et al. 2020). Our molecular findings further confirmed these outcomes, as SC upregulated hepatic *igf1* expression while downregulating growth-inhibitory genes *igfbp1a* and *mstn*, supporting its role in modulating endocrine pathways related to growth. These observations align with previous reports on various aquaculture species, including shrimp, tilapia, Seabream and trout (Abdel-Tawwab et al. 2020; Li Wang et al. 2018; Nimrat et al. 2021; El-Bab et al. 2022). Conversely, glyphosate exposure significantly impaired growth performance, which was associated with the downregulation of *igf1* and upregulation of *igfbp1a* and *myostatin*. This growth suppression is consistent with previous findings (Muhammad et al. 2021) and may be explained by GLY-induced damage to nutrient-absorbing and critical metabolic tissues such as the intestines, liver and gills, as observed histologically. Moreover, stress-related energy demands may have diverted nutrients away from growth to support detoxification and repair processes (Agbohessi et al. 2014).

The intestinal barrier, a key component in nutrient uptake and immune defence, was notably compromised in GLY-exposed fish. Histopathological damage included epithelial desquamation, villus blunting, crypt shortening and inflammation. In parallel, transcriptional profiling showed significant downregulation of tight junction genes (*ocln* and *cldn3*), indicative of disrupted epithelial integrity and increased intestinal permeability in the CON + GLY group. However, dietary SC supplementation preserved normal intestinal architecture and maintained or restored the expression of barrier-related genes. This protective effect is likely attributable to the functional components of yeast, particularly MOS and polyamines, which enhance epithelial regeneration, modulate microbial populations and support tight junctions (Sharma 2025). Similar protective effects of SC on gut barrier function have been documented in mice, broilers and piglets (Sivignon et al. 2015;

Che et al. 2017; W. Wang et al. 2016). These data underscore SC's capacity to mitigate herbicide-induced enteric damage and support intestinal homeostasis under toxicant stress.

Immune function was significantly disrupted by glyphosate exposure, as evidenced by downregulated expression of lysozyme and complement 3 (*lyz* and *c3*) and elevated expression of pro-inflammatory *tnfα*. These molecular alterations are indicative of immunosuppression and heightened inflammatory responses and are consistent with prior reports on glyphosate-induced immune dysfunction in various aquatic species (T. Zheng et al. 2021; Yousefi et al. 2021; Xiaozhen Yang et al. 2019). Glyphosate is known to disrupt immune responses through multiple mechanisms, including lipid peroxidation, suppression of immunoglobulin production, inhibition of immune cell proliferation and altered cytokine profiles (Abdelmagid et al. 2023). In contrast, dietary supplementation with SC markedly improved immune competence, as evidenced by restored expression of *lyz* and *c3* and reduced *tnfα* levels in both challenged and unchallenged fish. These effects may be attributed to the immunomodulatory action of yeast-derived β-glucans, which are known to activate phagocytosis, stimulate cytokine production, and enhance immune cell proliferation (El-Bab et al. 2022; W. Wang et al. 2016). In addition, the elevated lysozyme expression in SC-fed fish may also reflect enhanced lysosomal enzyme release and proliferation of phagocytes. These findings demonstrate the potential of SC to effectively mitigate glyphosate-induced immunotoxicity and restore immune function in Nile tilapia.

Biochemical blood parameters are widely recognized as reliable indicators of physiological stress and organ dysfunction in fish exposed to environmental pollutants including herbicides and pesticides (Ramaiah 2007; Bacchetta et al. 2014). In this study, glyphosate exposure significantly elevated serum levels of hepatic enzymes (AST and ALT), renal markers (creatinine and BUN) and lipid profile indicators (cholesterol, triglycerides and VLDL-C), reflecting hepatic and renal dysfunction as well as metabolic disturbance. These findings were supported by histopathological alterations in the liver, including vacuolation, necrosis, and sinusoidal congestion. Furthermore, glyphosate disrupted protein metabolism, as evidenced by reduced serum levels of total protein, albumin and globulin. Such disturbances are consistent with previous reports that link glyphosate and other pesticide exposure to oxidative stress-mediated hepatic damage due to the liver's role in xenobiotic detoxification and its vulnerability to redox imbalance (Mansour and Mossa 2009; Abdelmagid et al. 2022). Importantly, dietary supplementation with SC significantly mitigated these adverse biochemical alterations. SC restored serum levels of liver enzymes and improved protein profiles, confirming its hepatoprotective potential. These effects likely stem from the antioxidant, anti-inflammatory and nutrient-enriching properties of SC components such as β-glucans, nucleotides and MOS, which enhance hepatic resilience and functional integrity (Abd El-Naby et al. 2024; Hassaan et al. 2018). Similar protective effects have been observed with other functional supplements, such as Spirulina and β-glucans, in pesticide-challenged catfish (Mokhbatly et al. 2020). SC also led to a significant reduction in serum cholesterol, triglycerides and VLDL-C levels in both non-challenged and glyphosate-challenged fish. This aligns with prior studies where yeast supplementation improved lipid metabolism and nutrient assimilation in shrimp and tilapia (Ayiku et al. 2020; Abd El-Naby et al. 2024) Additionally, the nephroprotective effect of SC, evidenced by the reduction of creatinine and urea levels in the SC GLY group, corresponds with earlier findings in pesticide-exposed Nile tilapia supplemented with microalgae and β-glucans (Abdelhamid et al. 2020).

At the molecular level, glyphosate exposure triggered pronounced oxidative stress in Nile tilapia, as reflected by the downregulation of *nrf2* and *sod*, and the concurrent upregulation of *keap1*. Nrf2 is a key transcription factor that regulates antioxidant

defense by activating downstream genes including *sod*, *cat* and *gpx*, essential for neutralising reactive oxygen species (ROS) and maintaining redox homeostasis (J. F. Luo et al. 2018). The suppression of this pathway in the GLY-exposed group indicates a compromised antioxidative capacity, contributing to hepatic and systemic oxidative damage. SC supplementation effectively reversed these changes, upregulating *nrf2* and *sod*, while downregulating *keap1*, demonstrating its capacity to restore redox balance. This antioxidant effect, mediated by the activation of the *nrf2* signaling pathway, is likely driven by the bioactive components in yeast, including vitamins, trace elements, and antioxidant peptides, which are known to enhance endogenous antioxidant enzyme activities (P. Zhang et al. 2018; Z. Tao et al. 2023; Yuan et al. 2019). Beyond oxidative stress, inflammation and apoptosis are closely intertwined in glyphosate-induced toxicity. Elevated *tnfα* expression observed in the CON + GLY group underscores the activation of inflammatory signalling, which is known to exacerbate hepatic injury through NF-κB-mediated pathways (Tak and Firestein 2001; Jia et al. 2019a). SC-fed fish showed markedly reduced expression of *tnfα*, reflecting its anti-inflammatory potential. Yeast components, particularly β-glucans, are recognised for their immunomodulatory roles, including suppressing pro-inflammatory cytokines and enhancing mucosal barrier function (Martins et al. 2010; Foligné et al. 2010). Additionally, the expression of apoptosis-related genes *casp3* and *casp9* was significantly upregulated in GLY-exposed fish, indicating activation of the intrinsic apoptotic cascade via mitochondrial cytochrome c release and caspase activation (Jaeschke et al. 2018; Hao et al. 2019). SC supplementation attenuated these effects, reducing apoptotic gene expression and suggesting a protective mechanism against GLY-induced cell death. This anti-apoptotic effect may stem from improved mitochondrial integrity and inhibition of the caspase cascade, as observed in yeast-fed fish in prior toxicological models (L. Luo et al. 2017; W. Zhao et al. 2013).

Histopathological analyses further confirmed biochemical and molecular findings, revealing that glyphosate exposure caused pronounced tissue damage in Nile tilapia. Gills exhibited interlamellar epithelial hyperplasia, lamellar fusion, and mucous cell proliferation, hallmarks of impaired respiration and osmoregulation under toxicant stress as reported (Biagini et al. 2009; Pal et al. 2012). Liver tissues showed diffuse hepatocellular vacuolation, necrosis, pyknotic nuclei and sinusoidal congestion, indicating profound hepatic dysfunction consistent with pesticide-induced oxidative injury (Stentiford et al. 2003). Similar hepatic lesions have previously been reported in Nile tilapia exposed to glyphosate (Abdelmagid et al. 2022; Jiraungkoorskul et al. 2002). Intestinal tissues revealed villus blunting, crypt atrophy, epithelial desquamation, and lamina propria inflammation, compromising nutrient absorption and barrier integrity (Vajargah et al. 2021). SC supplementation significantly alleviated these lesions, preserving normal gill architecture, hepatic cellular morphology, and intestinal integrity, with markedly reduced inflammation and necrosis. These protective effects are likely due to the presence of MOS in yeast and its ability to produce polyamines, which help enhance health status by supporting mucosal integrity, reducing inflammation, and promoting cellular repair (El-Bab et al. 2022; Islam et al. 2021; Xuanyi Yang et al. 2020). Comparable results have been reported with dietary yeast, Spirulina, horseradish tree leaf extract, phytase and microalgae under toxicant exposure (Mokhbatly et al. 2020; Fadl et al. 2021; Elahwl et al. 2025), underscoring SC's efficacy in preserving organ health under glyphosate stress.

Conclusion

In conclusion, this study provides novel insights into the molecular and physiological mechanisms of glyphosate-induced toxicity in Nile tilapia, revealing significant impairments in growth, oxidative balance, immune function, and tissue integrity. Glyphosate exposure disrupted antioxidant pathways (*nrf2*, *sod*), upregulated pro-inflammatory (*tnfα*) and apoptotic (*cas3*, *cas9*) genes, altered lipid metabolism, and damaged hepatic, renal, intestinal, and gill structures. Dietary supplementation with Saccharomyces cerevisiae (SC) effectively alleviated these adverse effects by enhancing growth and feed efficiency, restoring antioxidant and immune responses, stabilising biochemical parameters, and preserving histological architecture. These protective effects are likely attributed to SC's bioactive constituents, including β-glucans, mannan oligosaccharides, vitamins, trace elements, and polyamines, which collectively support redox homeostasis, immune modulation, and tissue repair. Overall, the findings highlight the potential of SC as a functional feed additive to mitigate glyphosate-induced toxicity and promote resilience in aquaculture species.

Author contributions Z.I.E.: Conceptualization, Methodology, Supervision, Investigation, Writing-Review & Editing. A.S.S.: Methodology, Resources. I.A.: Conceptualization, Methodology, Formal analysis. A.E.: Writing-Original Draft, Methodology, Formal Analysis, Validation, Resources. M.A.: Investigation, Resources. A.A.: Investigation, Resources. D.H.A.: Writing-Original Draft, Investigation, Formal analysis, Resources.

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Data availability Data will be made available upon request.

Declarations

Ethics approval The experimental protocol was reviewed and approved by the Animal Ethics Committee of the Faculty of Aquatic and Fisheries Sciences, Kafrelsheikh University, Egypt (Approval No: IAACUC-KSU-3–2022).

Clinical trial number Not applicable.

Clinical trial registration Not applicable.

Uncropped gels and blots Not applicable as no gel or blot images are presented in the study.

Competing interests The authors declare no competing interests.

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