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RESEARCH

Mitigating deltamethrin-induced systemic toxicity in Nile Tilapia (*Oreochromis niloticus*) via dietary artichoke (*Cynara scolymus*) leaf extract: a multi-biomarker approach

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Abstract

This study investigated the potential of dietary artichoke (*Cynara scolymus*) leaf extract (ALE) to mitigate the multifaceted physiological disruptions induced by deltamethrin (DLM) exposure in Nile tilapia (*Oreochromis niloticus*). Following the determination of sublethal toxicity thresholds, fish were distributed into four experimental groups: a control, a DLM-exposed group (0.0014 mg/L), a group co-administered with DLM and 2.00% dietary ALE, and a group receiving ALE alone. Over a 30-day feeding trial, comprehensive assessments of growth kinetics, hematological profiles, serum biochemistry, and organ-scale histopathology were conducted. The results revealed that DLM exposure triggered significant growth retardation characterized by suppressed weight gain and impaired feed utilization ($p < 0.05$). Hematological indices such as RBC, Hb, and HCT were markedly reduced, while serum markers of hepatic stress encompassing glucose, cholesterol, and liver enzymes like ALT, AST, and ALP showed significant elevations in the DLM-only group. Furthermore, histopathological examinations confirmed DLM-induced structural damage within the liver, gills, and brain tissues. In contrast, dietary supplementation with 2.00% ALE significantly attenuated these toxicological impacts, supporting physiological balance and enhancing the antioxidant defense system (TAS, SOD, CAT, and GPx) while reducing lipid peroxidation (MDA). These findings suggest that *Cynara scolymus* leaf extract shows promise as a functional feed additive, providing significant mitigative potential against pyrethroid-induced oxidative stress and tissue degeneration in aquaculture systems.

Keywords *Cynara scolymus* · Deltamethrin · Functional feed · Hematology · Oxidative stress · Pyrethroid toxicity · Nile tilapia

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Introduction

The rapidly increasing demand for food worldwide increases the importance of the aquaculture sector, which is the most sustainable production model among animal protein sources. Aquaculture not only involves the production of marine and freshwater products, but also has a strategic position in terms of economic development, food security and employment. The aquaculture sector is one of the fastest growing food sectors (FAO 2022).

Nile tilapia (*Oreochromis niloticus*) is an important freshwater species widely cultivated in both developed and developing countries due to its fast growth rate, resistance to environmental stresses, low feed cost and high market demand (El-Sayed 2006; Çelik et al. 2024). Global Nile tilapia production reached 5.3 million tonnes in 2022, making it the second largest aquacultured fish species after grass carp (FAO 2024).

Since open water resources used in traditional aquaculture systems are often susceptible to chemical pollution, particularly from surface runoff associated with agricultural activities, cultivated species may be exposed to pesticide residues. Deltamethrin, as a pyrethroid group pesticide, is widely used in agricultural areas and can have toxic effects on aquatic organisms when transported to surface waters (Köprücü and Aydın, 2004; Gewaily et al. 2021). Due to its lipophilic nature and limited solubility in environmental water, deltamethrin accumulates in fish tissues and causes multifaceted physiopathological disorders such as oxidative stress, growth retardation, hematological changes and histological damage (Amin and Hashem 2012; Ullah et al 2019). The basic mechanism of oxidative stress in fish is explained by an increase in the production of reactive oxygen species (ROS) in the body and a decrease in the functions of antioxidant enzymes such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx) involved in the detoxification of these species. Disruption of this balance leads to an increase in malondialdehyde (MDA) levels by increasing lipid peroxidation that causes damage to cell membranes (El-Sayed and Saad 2008).

The use of feed additives can be an effective way to tackle the challenges faced in aquaculture (Öz, 2018, 2019; Ahmadifar et al. 2021; Öz et al. 2018; Öz and Inanan 2025). To increase fish production and maintain fish welfare, various natural plant additives are used in the aquaculture industry to promote fish growth, immune function and antioxidant capacity (Inanan et al. 2021; Ahmadifar et al. 2021; Acar et al. 2019). Dietary supplementation with functional plant-based additives may offer alternative and natural solutions to mitigate the damages caused by waterborne pesticides (Tok et al. 2025; Öz et al. 2024a, 2024b). Artichoke (*Cynara scolymus*) leaf extract is a natural additive with high antioxidant capacity thanks to its chlorogenic acid, flavonoids and phenolic compounds (Vamanu et al. 2011). This extract stands out with its anti-inflammatory, hepatoprotective and immunosupportive properties and offers potential benefits in protecting fish health (Esen et al. 2025; Shohreh et al. 2025; Esen 2024; Elsayyad et al. 2024; Salem et al. 2017).

Artichoke (*Cynara scolymus*) is a well-recognized source of natural bioactive compounds, particularly phenolic acids and flavonoids. The biological activity of artichoke leaf extract (ALE) is mainly attributed to its characteristic phenolic constituents, including caffeoylquinic acids such as cynarin (1,3-dicaffeoylquinic acid) and chlorogenic acid (5-caffeoylquinic acid), as well as flavonoids such as luteolin and apigenin. These compounds exhibit strong antioxidant, hepatoprotective, and anti-inflammatory properties through effective scavenging of reactive oxygen species (ROS) and modulation of endogenous antioxidant defense systems (Ayuso et al. 2024; Gil-Martínez et al. 2025; Zuccolo

et al. 2025). In the present study, a standardized commercial aqueous artichoke leaf extract was used to evaluate its protective efficacy against pesticide-induced toxicity in Nile tilapia.

Consequently, the present study was designed to comprehensively evaluate the mitigative potential of dietary artichoke leaf extract against the systemic toxicity induced by waterborne deltamethrin in Nile tilapia. By focusing on a multi-dimensional analysis of growth kinetics, hemato-biochemical profiles, and oxidative stress markers, alongside detailed histopathological assessments of vital organs, this research seeks to elucidate the protective mechanisms of *Cynara scolymus*. The findings are expected to provide critical insights into the integration of bioactive plant extracts as functional feed ingredients, ultimately contributing to the development of sustainable nutritional strategies aimed at enhancing fish health and resilience in pesticide-impacted aquaculture systems.

While several studies have explored botanical mitigants, the novelty of this research lies in its holistic multi-biomarker integration, specifically examining the protective role of *Cynara scolymus* against neuro and muscular tissue degradation areas that remain underinvestigated in Nile tilapia toxicity models.

Materials and methods

Fish material and experimental design

This research was carried out within the framework of the ethical approval obtained from the "Çukurova University Aquatic Vertebrates Experiments Local Ethics Committee on 07.05.2024 and in accordance with the principles of the Local Ethics Committee. In this research, 240 Nile tilapia with an average live weight of 87.66 ± 2.08 g obtained from Çukurova University Faculty of Fisheries Dr. Nazmi Tekelioğlu Freshwater Fisheries Production and Research Station were used as subjects. The experimental part of our research consisted of two stages. Analytical-standard deltamethrin (PESTANAL®, Sigma-Aldrich, CAS 52918–63-5; neat) was used to prepare stock solutions for exposure experiments. The primary stock solution was prepared by dissolving 140 mg of deltamethrin in 10 mL of 100% acetone to achieve a concentration of 14 mg/mL. This stock was subsequently diluted with aerated water to achieve the desired exposure concentrations. The final acetone concentration in all exposure tanks was $< 0.01\%$ (v/v), a level widely considered non-toxic to fish and consistent with standard aquatic toxicity testing protocols (OECD 2025). Consequently, a separate solvent control group was not deemed necessary as this concentration remained uniform across all groups receiving the toxicant. To determine the concentration of deltamethrin for the exposure trial, the 96-h LC50 value was first determined to be 0.028 mg/L using the probit analysis method. For this purpose, 120 fish subjects were distributed among six different concentrations (0.00, 0.01, 0.02, 0.04, 0.08, and 0.16 mg/L) of deltamethrin. The subsequent feeding trial was planned using deltamethrin at a rate of 1/20 of this calculated LC_{50} value. The LC_{50} test was conducted using a static renewal system in 50 L aquariums. To maintain stable concentrations and minimize photodegradation, the water was replaced every 24 h using stock solutions stored in light-protected containers. Throughout the 96-h period, the tanks were continuously aerated, and the subjects were monitored three times daily for mortality, with any dead fish promptly removed. Fish were not fed during this acute exposure phase.

The study followed a completely randomized design (CRD), where fish were randomly allocated to treatment groups with three replicate tanks per treatment. The feeding phase was conducted in 80 L aquariums, each containing 10 fish and was carried out in triplicate. A total of 30 fish were used for each experimental group. Water temperature was maintained at a constant 25 °C using Eheim 100 W thermostat heaters in all aquariums. Experimental groups were designated as G1 (control), G2 (artichoke), G3 (deltamethrin), and G4 (deltamethrin + artichoke), as summarized in Table 1. Subjects selected for hematological, serum biochemical, oxidative stress, and histopathological analyses were sampled from all replicate tanks in each treatment group to eliminate tank-related bias.

Preparation of trial feeds and feeding protocol

The basal diet consisted of commercially pelleted tilapia feed (Hem Yem, Gaziantep, Turkey) containing 39% crude protein, 6.7% crude lipid, 4.30% crude fiber, and 6.79% crude ash on a dry matter basis. This feed was used as the control diet and as the base for the experimental diets supplemented with artichoke leaf extract. Artichoke leaf aqueous extract used in the experiment was supplied by a commercial company (Immunat; TR-48-K-019618). In the study, 2.00% artichoke leaf aqueous extract was added to the fish feed by spraying method (Esen 2024). Experimental feeds were prepared in batches of 200 g, and the extract was homogeneously applied to the pellets by a spraying method. To ensure consistency across all groups, the control diets (G1 and G3) were similarly sprayed with an equivalent volume of distilled water (devoid of the extract), while the ALE-supplemented diets (G2 and G4) received the 2.00% artichoke leaf aqueous extract. The extract (or water for the control) was sprayed onto the feed pellets while manually mixing the batch to ensure uniform coating, followed by air-drying at room temperature before feeding. Given the minor inclusion level (2.00%) of the extract, the proximate composition of the basal diet remained statistically consistent across all experimental groups, ensuring that the observed effects were not influenced by variations in macronutrient intake. To ensure the stability of the bioactive phenolic compounds within the ALE, the air-dried experimental diets were stored in airtight, light-protected containers at 4 °C throughout the 30-day trial. While batch-specific chemical characterization of the ALE was not performed locally, the study utilized a standardized commercial extract with a consistent bioactive profile guaranteed by the manufacturer to contain key markers such as cynarin and chlorogenic acid to ensure experimental reproducibility.

The fish placed in the experimental tanks in the research unit were re-weighed after the completion of the 15-day adaptation period and started to be fed with the experimental feeds the next day. At the beginning of the experiment, the fish were fed a fixed ration of 2% of their body weight to ensure standardized intake and environmental stability during

Table 1 Experimental design and treatment groups showing dietary artichoke supplementation and deltamethrin exposure levels in Nile tilapia

Groups	Extract in feed (%)	Deltamethrin in water (mg/L)	Number of fish
G1	0.00	0.00	30 (3*10)
G2	2.00	0.00	30 (3*10)
G3	0.00	$LC_{50}/20$	30 (3*10)
G4	2.00	$LC_{50}/20$	30 (3*10)
Total			**120**

the initial adaptation phase. From the second week onwards, the protocol was transitioned to feeding to visual satiety (ad libitum) to allow for the assessment of maximum growth potential and voluntary feed intake under the experimental conditions. Throughout the 30-day trial, the fish were fed two meals daily at 09:30 and 18:00.

Water temperatures were checked regularly in the morning and evening using digital thermometers placed in the aquariums. In addition, the airstones were inspected regularly to ensure homogeneous distribution of aeration throughout the tanks. Dissolved oxygen was maintained between 6.5–7.5 mg/L by continuous aeration and measured using an OxyGuard® oxygen meter once daily after feeding. The pH of the rearing water ranged from 7.3 to 7.8. Overall, water quality parameters were monitored regularly to ensure stable experimental conditions.

Measurement of fish growth parameters

Initial weight (IW), intermediate measurements and final weight (FW) measurements were made individually with a KERR brand precision balance sensitive to 0.1 g and the following calculations were used to evaluate growth performance and feed data in the experiment.

Specific Growth Rate (SGR) = [(ln FW − ln IW)/number of days] × 100, (Company et al 1999)

$$\text{Live weight gain (LWG, g)} = \text{Final Weight (FW)} - \text{Initial Weight (IW)}$$

$$\text{Daily feed intake (DFI)} = \text{Amount of feed consumed/time}$$

Feed conversion ratio (FCR) = Amount of feed consumed/weight gain (Santinha et al 1999).

Protein efficiency ratio (PER) = Live weight gain (g)/protein intake (g), (Skalli and Robin 2004).

Blood sampling and measurement of hematologic parameters

Fish subjects were fasted for 24 h prior to blood sampling to avoid post-prandial effects on hematological and biochemical parameters. Blood samples were collected from the caudal vein of the fish using a 1 mL syringe equipped with a 30G needle. The collected samples were immediately transferred into 0.5 mL blood tubes containing K_3EDTA for hematological analysis or into tubes without anticoagulant for serum separation. Samples in the anticoagulant-free tubes were centrifuged in a refrigerated centrifuge at 1200 X g for 15 min to obtain serum, which was then stored at −80 °C for subsequent biochemical and oxidative stress analyses.

Hematological analyses were performed at Aksaray University Embryo Transfer Center using an MS4S brand blood counting device. Before the analysis, the results obtained by the device for Nile tilapia blood were compared with data obtained by the manual method (Blaxhall and Daisley 1973) and the device was calibrated. The integration of automated hematological analysis and the use of standardized blood indices for Nile tilapia were performed according to established protocols to ensure methodological accuracy. Furthermore, the validation of these hematological parameters followed the comprehensive framework described by Fazio (2019). Within the scope of these analyses, parameters including red blood cell (RBC), hemoglobin (Hb), hematocrit (Hct), mean red blood cell volume

(MCV), mean red blood cell hemoglobin (MCH), and mean red blood cell hemoglobin concentration (MCHC) were evaluated. To ensure methodological rigor and adhere to the experimental unit (tank), hematological analyses were performed on samples from nine fish per treatment (three fish per replicate tank), and the results were averaged to represent tank-level replicates (n = 3 per group) prior to statistical analysis.

Determination of biochemistry parameters

ALP (alkaline phosphatase), AST (aspartate aminotransferase), ALT (alanine aminotransferase), TP (total protein), TRG (triglyceride), Cho (cholesterol), and Glu (glucose) were analyzed as serum biochemical parameters. An automatic biochemistry device (MINDRAY-BS400) was used for the analysis of biochemical parameters (Tok et al. 2025). To ensure consistency with the experimental unit, biochemical indices were measured from nine fish per treatment (three fish per replicate tank), and the values were averaged to represent tank-level replicates ($n = 3$ per group) prior to statistical analysis.

Oxidative stress parameters

Sera extracted from blood samples of fish were analyzed for TAS; Total Antioxidant Status, TOS; Total Oxidant Status, SOD; SuperOxide Dismutase, CAT; Catalase, GPx; Glutathione Peroxidase and MDA; Malondialdehyde.

TAS (Total Antioxidant Status) levels were measured using Relassay (Cat no: RL0017) brand commercial kits (Erel 2004). TOS (Total Oxidant Status) levels were evaluated using Relassay (Cat no: RL0024) brand commercial kits (Erel 2005). The ratio of TOS to TAS is considered as oxidative stress index (OSI). The TAS unit obtained for calculation was converted to µmol/L and the OSI value was calculated according to the following formula.

OSI = TOS (µmol H_2O_2 equivalent/L)/TAS (µmol Trolox equivalent/L) (Yumru et al 2009; Kösecik et al. 2005; Harma et al. 2003). Oxidative stress index (OSI) was calculated as the ratio of total oxidant status (TOS) to total antioxidant capacity (TAC) and expressed as arbitrary units (AU). Although some studies express OSI as a percentage (× 100), the present study used the direct ratio approach, as both methods are mathematically equivalent.

Malondialdehyde (MDA) level, a product of lipid peroxidation, was determined according to the study (Alak et al. 2020). Superoxide dismutase (SOD) enzyme activity was determined spectrophotometrically at 560 nm wavelength using the method based on the reduction reaction between superoxide anions (O_2-) and nitro blue tetrazolium (NBT) under light (Sun et al. 1988). Measurement of catalase (CAT) activity; samples were mixed with 1 mL H_2O_2 (50 mM) and reacted at 37 °C for 1 min. Then 1 mL of ammonium molybdate was added to terminate the reaction, resulting in the formation of a yellowish complex containing H_2O_2 residue. Finally, the UV-visabsorption of this complex was measured at 405 nm by a microplate reader (Cheng et al. 2020).

Measurement of glutathione peroxidase (GPx) activity; Glutathione Peroxidase (GPx) catalyzes the oxidation of glutathione by cumene hydroperoxide. In the presence of glutathione (GSSG), it is immediately converted to the reduced form by simultaneous oxidation of NADPH to NADP. GPx activity was measured by a change in absorbance (decrease in readings over 3 min) at 340 nm (Paglia and Valentine 1967; Prohaska et al. 1977; Kraus and Ganther 1980).

Oxidative stress parameters were determined from serum samples collected from nine fish per experimental group (three fish per replicate tank). To maintain consistency with the experimental unit, the values from fish within the same tank were averaged, and all statistical comparisons were performed based on tank-level replicates (n = 3 per group).

Histopathological methods

Following euthanasia, specific tissue samples were collected from both the right and left second gill arches, the central liver lobe, the whole brain, and the dorsal muscle (epaxial region) of each fish. These tissues were fixed in 0.1 M phosphate-buffered formaldehyde solution (pH 7.4). After trimming the tissue edges, the samples were washed under slowly flowing tap water for 24 h. Subsequently, dehydration was performed through an ascending ethanol series (70%, 80%, 90%, 96%, and 100%) for 60 min at each step. The tissues were then cleared in xylene and infiltrated with xylene-paraffin for 30 min each. Prior to embedding, the samples were sequentially kept in soft paraffin (46–48 °C) for 15 min and hard paraffin (56–58 °C) for 30 min using a Leica EG 1150 H embedding station. Sections with a thickness of 4 µm were obtained using a Leica RM2125 Rotary Microtome. These sections were stained with the Hematoxylin–Eosin (H&E) technique, dehydrated, cleared, and mounted on glass slides using Entellan (Merck). Microscopic examination was conducted with a Leica DM-750 light microscope, and digital images of the identified lesions were recorded according to the criteria of Culling et al. (2014). To ensure unbiased assessment, slides were blindly coded and examined by the pathologist without knowledge of the treatment groups. For each experimental group, tissue samples from nine fish were processed, and the findings from three fish per replicate tank were evaluated to represent the tank-level histological profile ($n = 3$ per group).

Statistical analysis

Prior to statistical analysis, data were tested for normality using the Kolmogorov–Smirnov test and for homogeneity of variances using Levene's test. A two-way analysis of variance (ANOVA) was employed to evaluate the main effects of dietary artichoke leaf extract (ALE; 0% and 2%) and deltamethrin (Delta; 0 and $LC_{50}/20$), as well as their interaction (ALE × Delta). When a significant interaction was detected ($p < 0.05$), differences among treatment combinations were evaluated using Tukey's HSD post-hoc test. For variables without a significant interaction, the main effects were interpreted independently to avoid overinterpretation of biological interactions. The significance level was set at $p < 0.05$. The experiment followed a completely randomized design with three replicate tanks per treatment, and the tank was considered the experimental unit. To ensure methodological rigor and strictly avoid pseudo-replication, data for the three fish sampled from each replicate tank (for hematology, biochemistry, and oxidative stress) were averaged prior to statistical analysis. Consequently, all parameters were analyzed using tank-level replication (n = 3 per treatment).

Results and discussion

Results

Growth parameters of fish

Two-way ANOVA revealed significant main effects of both dietary artichoke leaf extract (ALE) and deltamethrin exposure on all growth performance parameters ($p < 0.001$; Table 2).

Deltamethrin exposure alone (G3) induced significant growth retardation characterized by reduced weight gain, specific growth rate (SGR), and feed utilization efficiency compared with the control group ($p < 0.05$). A significant ALE × Delta interaction was detected specifically for feed utilization indices (FCR and PER), indicating that the protective effect of dietary ALE on feed efficiency was statistically modulated by the presence of toxic stress ($p < 0.05$). In contrast, for parameters such as weight gain and SGR, where no significant interaction was observed ($p > 0.05$), both ALE and deltamethrin exerted independent main effects. Accordingly, fish receiving 2% ALE under deltamethrin exposure (G4) exhibited significantly improved growth performance relative to G3. Notably, ALE supplementation not only mitigated deltamethrin-induced growth suppression but also effectively restored the final weight and growth parameters to levels that met and significantly exceeded those of the Control group (G1), demonstrating a complete physiological recovery and a growth-promoting effect ($p < 0.05$).

Hematological parameters of fish

The hematological profiles of Nile tilapia (*Oreochromis niloticus*) following the 30-day growth trial are summarized in Table 3. Two-way ANOVA revealed that dietary ALE

Table 2 Growth performance of Nile tilapia (*Oreochromis niloticus*) fed diets supplemented with artichoke leaf extract (ALE) under deltamethrin (Delta) exposure

Parameter	G1 (Control)	G2 (ALE)	G3 (Delta)	G4 (ALE + Delta)	*p* (ALE)	*p* (Delta)	*p* (Int.)
IW	87.66 ± 2.08	87.66 ± 2.08	87.66 ± 2.08	87.66 ± 2.08			
FW (g)	126.81 ± 1.15^{c}	141.52 ± 0.76^{a}	116.61 ± 1.07^{d}	131.95 ± 0.88^{b}	< 0.001	< 0.001	0.596
WG (g)	39.15 ± 1.15^{c}	53.86 ± 0.76^{a}	28.95 ± 1.07^{d}	44.29 ± 0.88^{b}	< 0.001	< 0.001	0.596
FI (g)	70.17 ± 1.59^{c}	84.73 ± 1.10^{a}	64.34 ± 0.57^{d}	76.13 ± 0.98^{b}	< 0.001	< 0.001	0.065
DFI (g/day)	2.34 ± 0.05^{c}	2.82 ± 0.04^{a}	2.14 ± 0.02^{d}	2.54 ± 0.03^{b}	< 0.001	< 0.001	0.065
FCR	1.79 ± 0.01^{b}	1.57 ± 0.04^{c}	2.22 ± 0.10^{a}	1.72 ± 0.05^{b}	< 0.001	< 0.001	0.003
SGR (%)	1.23 ± 0.03^{c}	1.60 ± 0.02^{a}	0.95 ± 0.03^{d}	1.36 ± 0.02^{b}	< 0.001	< 0.001	0.162
PER	1.43 ± 0.01^{b}	1.63 ± 0.04^{a}	1.15 ± 0.05^{c}	1.49 ± 0.04^{b}	< 0.001	< 0.001	0.019

IW Initial fish weight, *FW* Final weight, *FI* Feed intake, *DFI* Daily feed intake, *FCR* Feed conversion rates, *SGR* Specific growth rate, *PER* Protein efficiency ratio. The experiment was conducted using a completely randomized design with three replicate tanks per treatment. Values are presented as mean ± SD ($n = 3$ replicates per group). Different superscript letters (a–d) within the same row indicate significant differences among groups ($p < 0.05$; two-way ANOVA followed by Tukey's HSD test). *p* (ALE): main effect of dietary artichoke extract; *p* (Delta): main effect of deltamethrin exposure; p (Int.): interaction effect between ALE and deltamethrin

Table 3 Hematological responses of Nile tilapia (*Oreochromis niloticus*) fed artichoke leaf extract (ALE) supplemented diets under deltamethrin (Delta) exposure

Parameters	G1 (Control)	G2 (ALE)	G3 (Delta)	G4 (ALE + Delta)	*p* (ALE)	p (Delta)	*p* (Int.)
RBC (10^6/mm^3)	1.62 ± 0.25^{b}	1.97 ± 0.02^{a}	1.43 ± 0.01^{c}	1.82 ± 0.01^{b}	< 0.001	0.015	0.623
Hb (g/dl)	12.67 ± 0.38^{c}	16.93 ± 0.41^{a}	10.88 ± 0.31^{d}	14.33 ± 0.26^{b}	< 0.001	< 0.001	0.010
Hct (%)	41.99 ± 0.46^{c}	52.87 ± 0.28^{a}	40.52 ± 1.18^{d}	44.22 ± 0.66^{b}	< 0.001	0.002	< 0.001
MCV (fl)	246.51 ± 9.99^{b}	269.51 ± 3.49^{a}	284.02 ± 8.97^{a}	242.50 ± 2.39^{b}	0.003	0.067	< 0.001
MCH (pg)	74.35 ± 3.23^{c}	86.10 ± 1.96^{a}	76.28 ± 2.01^{bc}	78.62 ± 1.65^{b}	< 0.001	0.007	< 0.001
MCHC (g/dl)	30.17 ± 0.98^{b}	32.03 ± 0.92^{a}	26.88 ± 1.10^{c}	32.42 ± 0.79^{a}	0.003	< 0.001	< 0.001

Values are presented as mean ± SD ($n = 3$ tank-level replicates per group)**.** Data from the three fish sampled per replicate tank were averaged prior to statistical analysis to ensure the tank remained the experimental unit. Different superscript letters (a–d) within the same row indicate significant differences among groups ($p < 0.05$; two-way ANOVA followed by Tukey's HSD test). *p* (ALE): main effect of artichoke; *p* (Delta): main effect of deltamethrin; p (Int.): interaction effect

supplementation and deltamethrin (DLM) exposure significantly influenced the hematological status of the fish ($p < 0.05$). Highly significant interaction effects (pInt < 0.01) were observed for Hb, Hct, MCV, MCH, and MCHC, statistically confirming that the mitigative efficacy of ALE on these parameters was specifically modulated by the presence of pesticide-induced stress. In contrast, for RBC count, where the interaction effect was non-significant ($p = 0.623$), dietary ALE and DLM exposure exerted independent main effects.

A marked reduction in RBC ($1.43 \pm 0.01 \times 10^6$/mm^3), Hb ($10.88 \pm 0.31$ g/dL), and Hct ($40.52 \pm 1.18\%$) was recorded in the DLM-only group (G3), indicating a suppressed hematopoietic system. While ALE supplementation in group G4 led to an overall improvement in RBC levels via its independent main effect, the significant recovery in Hb and Hct values toward control levels (14.33 ± 0.26 g/dL and $44.22 \pm 0.66\%$, respectively) was statistically supported by a significant ALE × DLM interaction. Furthermore, erythrocyte indices were significantly altered by DLM exposure; the G3 group exhibited the highest MCV and the lowest MCHC values ($p < 0.05$). These disturbances were markedly neutralized in the ALE-supplemented groups, particularly in G4, where indices remained comparable to the control (G1). These results demonstrate that the bioactive phenolic compounds in artichoke leaf extract stabilize erythrocyte membrane integrity and counteract the hemolytic impacts of deltamethrin.

Oxidative stress parameters of fish

The oxidative stress parameters of Nile tilapia (*Oreochromis niloticus*) at the conclusion of the 30-day growth trial are presented in Table 4. Two-way ANOVA revealed that both dietary ALE supplementation and deltamethrin exposure significantly influenced the oxidative status of the fish ($p < 0.001$). Notably, significant interaction effects ($p < 0.05$) were detected for MDA, TOS, OSI, CAT, and GPx, statistically confirming that the mitigative efficacy of ALE against deltamethrin-induced oxidative damage was specifically modulated by the treatment combination. Conversely, for TAS and SOD activity, where no significant interaction was observed ($p > 0.05$), both ALE and deltamethrin exerted independent main effects on the antioxidant profile.

Table 4 Oxidative stress responses of Nile tilapia (*Oreochromis niloticus*) fed artichoke leaf extract (ALE) supplemented diets under deltamethrin (Delta) exposure

Parameters	G1 (Control)	G2 (ALE)	G3 (Delta)	G4 (ALE + Delta)	*p* (ALE)	*p* (Delta)	*p* (Int.)
TAS (mmol/L)	2.64 ± 0.03^{c}	3.04 ± 0.05^{a}	2.41 ± 0.02^{d}	2.82 ± 0.03^{b}	< 0.001	< 0.001	0.562
TOS (µmol/L)	18.79 ± 0.31^{c}	28.28 ± 0.43^{a}	16.01 ± 0.27^{d}	23.50 ± 0.23^{b}	< 0.001	< 0.001	< 0.001
OSI	0.71 ± 0.01^{c}	0.93 ± 0.01^{a}	0.66 ± 0.01^{d}	0.83 ± 0.01^{b}	< 0.001	< 0.001	< 0.001
CAT (U/L)	209.55 ± 1.37^{c}	252.57 ± 3.25^{a}	162.89 ± 3.25^{d}	231.80 ± 2.63^{b}	< 0.001	< 0.001	0.003
SOD (U/L)	428.69 ± 2.66^{c}	474.25 ± 2.95^{a}	403.89 ± 2.53^{d}	452.27 ± 2.23^{b}	< 0.001	< 0.001	0.091
MDA (µmol/L)	94.51 ± 0.77^{b}	85.36 ± 1.05^{d}	109.06 ± 0.72^{a}	91.65 ± 0.71^{c}	< 0.001	< 0.001	< 0.001
GPx (U/L)	189.38 ± 0.84^{c}	242.82 ± 0.53^{a}	161.89 ± 2.01^{d}	214.24 ± 1.04^{b}	< 0.001	< 0.001	< 0.001

Values are presented as mean ± SD ($n = 3$ tank-level replicates per group). To ensure the tank remained the experimental unit, data from the three fish sampled per replicate were averaged prior to statistical analysis. Different superscript letters (a,b,c,d) within the same row indicate significant differences among groups ($p < 0.05$; Two-way ANOVA followed by Tukey's HSD test). *p* (ALE): effect of artichoke; *p* (Delta): effect of deltamethrin; *p* (Int.): interaction effect. *TAS* Total Antioxidant Status, *TOS* Total Oxidant Status, *OSI* Oxidative Stress Index, *CAT* Catalase, *SOD* Superoxide Dismutase, *MDA* Malondialdehyde, *GPx* Glutathione Peroxidase

Significant changes were recorded in the oxidative stress markers of the deltamethrin-treated G3 group. While antioxidant enzyme activities (SOD, CAT, and GPx) and total antioxidant status (TAS) decreased significantly in the G3 group, malondialdehyde (MDA) levels exhibited a significant increase ($p < 0.05$), indicating increased lipid peroxidation and a disruption in oxidative balance. Interestingly, total oxidant status (TOS) and the oxidative stress index (OSI) were found to be lower in the G3 group compared to the control.

In contrast, the oxidative stress profile was significantly improved in the ALE-supplemented G4 group. In this group, the significant recovery in CAT and GPx activities, along with the reduction in MDA levels compared to the G3 group, was statistically supported by a significant interaction effect ($p < 0.05$). While TAS and SOD levels also showed improvement in the G4 group, these shifts were primarily driven by the independent main effects of the extract and the toxicant. These findings demonstrate that artichoke extract modulates the antioxidant defense system to counteract the systemic oxidative stress impacts induced by deltamethrin exposure.

Serum biochemistry parameters of fish

The serum biochemical profiles of Nile tilapia (*Oreochromis niloticus*) were significantly influenced by both dietary ALE supplementation and deltamethrin exposure ($p < 0.001$; Table 5). According to the two-way ANOVA results, a significant interaction ($p < 0.05$) was detected for almost all parameters, including ALP, AST, glucose, cholesterol, total protein, and triglycerides, demonstrating that ALE specifically modulated the deltamethrin-induced metabolic and hepatic disruptions. However, for ALT activity, no significant interaction was observed ($p = 0.849$), indicating that ALE and deltamethrin exerted independent main effects on this specific liver enzyme.

Exposure to deltamethrin alone (G3) led to a marked elevation in liver enzyme activities (AST, ALT, and ALP), which serve as critical indicators of hepatic stress and cellular

Table 5 Serum biochemical profiles of Nile tilapia (*Oreochromis niloticus*) fed artichoke leaf extract (ALE) supplemented diets under deltamethrin (Delta) exposure

Parameters	G1 (Control)	G2 (ALE)	G3 (Delta)	G4 (ALE + Delta)	*p* (ALE)	*p* (Delta)	*p* (Int.)
ALP (U/L)	61.04 ± 1.87^{c}	42.06 ± 0.23^{d}	86.41 ± 1.09^{a}	52.78 ± 0.51^{b}	< 0.001	< 0.001	< 0.001
AST (U/L)	290.63 ± 1.36^{c}	255.84 ± 0.55^{d}	330.65 ± 0.75^{a}	269.92 ± 1.59^{b}	< 0.001	< 0.001	< 0.001
ALT (U/L)	35.17 ± 1.03^{b}	17.43 ± 0.53^{c}	41.11 ± 0.29^{a}	23.22 ± 0.35^{c}	< 0.001	< 0.001	0.849
Glu (mg/dl)	44.34 ± 0.80^{b}	32.50 ± 0.50^{d}	54.41 ± 1.06^{a}	37.80 ± 0.83^{c}	< 0.001	< 0.001	0.001
Cho (mg/dl)	145.67 ± 0.94^{b}	116.73 ± 0.49^{d}	163.01 ± 0.74^{a}	131.02 ± 1.17^{c}	< 0.001	< 0.001	0.016
TP (g/dl)	6.86 ± 0.11^{b}	7.66 ± 0.08^{a}	4.91 ± 0.02^{d}	5.43 ± 0.02^{c}	< 0.001	< 0.001	0.009
Trg (mg/dl)	114.77 ± 2.37^{b}	71.07 ± 1.51^{d}	156.94 ± 1.28^{a}	91.63 ± 1.55^{c}	< 0.001	< 0.001	< 0.001

Values are presented as mean ± SD ($n = 3$ tank-level replicates per group). Data from the three fish sampled per replicate tank were averaged prior to statistical analysis to ensure the tank remained the experimental unit. Different superscript letters (a,b,c,d) within the same row indicate significant differences among groups ($p < 0.05$; Two-way ANOVA followed by Tukey's HSD test). p (ALE): main effect of artichoke; p (Delta): main effect of deltamethrin; p (Int.): interaction effect. *ALP* Alkaline phosphatase, *AST* Aspartate aminotransferase, *ALT* Alanine aminotransferase, *TP* Total protein, *TRG* Triglyceride, *Cho* Cholesterol, *Trg* Triglyceride, *Glu* Glucose

damage. Additionally, significant hyperglycemic (Glucose) and hyperlipidemic (Cholesterol and Triglyceride) responses were recorded in the G3 group, alongside a substantial reduction in total protein (TP) levels. In contrast, dietary supplementation with 2% artichoke leaf extract (G4) significantly attenuated these biochemical impairments. In the G4 group, the significant lowering of AST, ALP, glucose, and cholesterol levels toward control values was statistically supported by a significant ALE × Delta interaction. While ALT levels were also effectively reduced in the G4 group, this improvement was driven by the independent main effects of the extract and the toxicant rather than a synergistic interaction. These findings statistically confirm the potent hepatoprotective and metabolic-regulatory efficacy of *Cynara scolymus* leaf extract against pyrethroid-induced systemic toxicity.

Histopathological findings

Histopathological alterations were assessed qualitatively based on lesion presence and severity across experimental groups (Table 6). Light microscopic evaluations revealed distinct structural alterations in the gills, liver, brain, and muscle tissues of Nile tilapia, with the most pronounced damage occurring in the gill and hepatic architectures. In the control group (G1), the primary and secondary lamellae exhibited a standard histological organization with no detectable lesions. However, deltamethrin exposure in Group 3 (G3) induced severe pathological transformations, including epithelial desquamation, extensive edema, hyperplasia, and lamellar fusion. While Group 2 (G2) showed a predominantly normal structure with only negligible alterations, the ALE-supplemented group (G4) demonstrated a significant restoration of gill integrity. The gills in Group 4 displayed only minimal lesions, maintaining a structural appearance closely resembling the control group.

The hepatic tissue of G1 fish was characterized by normal hepatocyte morphology, well-defined sinusoidal spaces, and intact exocrine pancreatic acini. In contrast, G3 exhibited severe hepatotoxicity manifested by extensive hydropic and vacuolar degeneration, dilation of sinusoidal spaces, and significant focal hemorrhages. While G2 fish presented a regular histological profile with only slight sinusoidal congestion, the administration of ALE in G4

Table 6 Semiquantitative histopathological scoring of fish tissues in Nile tilapia (*Oreochromis niloticus*) fed 2% artichoke (*Cynara scolymus*) leaf extract (ALE) and/or exposed to sublethal deltamethrin (DLM; $LC_{50}/20$)

Tissue	Histopathological Alteration	G1 (Control)	G2 (ALE)	G3 (Delta)	G4 (ALE + Delta)
Gill	Separation of epithelial cells of the secondary lamellae	0	0–1	2–3	2
	Fusion of secondary lamellae	0	0–1	3	1–2
	Edema in secondary lamellae	0–1	1–2	2–3	1
Liver	Vacuolar degeneration	0	1–2	2	1–2
	Hydropic degeneration	0	0–1	2–3	2
	Congestion	0–1	1–2	3	1–2
Muscle	Atrophy	0–1	1–2	3	2
Brain	Hyperemia	0	1–2	2	1–2
	Vacuolization	0–1	1–2	2–3	1–2

Tissue sections were stained with Hematoxylin–Eosin (H&E) and examined under light microscopy. For each experimental group, tissue samples from nine fish were processed, and the findings from three fish per replicate tank were evaluated to represent the tank-level histological profile ($n = 3$ per treatment). Scoring criteria: 0 = absent; 1 = mild; 2 = moderate; 3 = severe

notably mitigated these deltamethrin-induced impairments. The liver sections in Group 4 exhibited minimal histopathological damage, confirming the hepatoprotective efficacy of artichoke extract, which aligns with recent findings on the protective roles of natural additives in tilapia.

Muscle tissues in G1, G2, and G4 maintained a regular histological arrangement with intact fibers. However, G3 exhibited noticeable muscle atrophy, a typical sign of metabolic stress induced by toxicant exposure. Similarly, the brain tissue in the control group appeared histologically normal. Deltamethrin exposure in G3 led to the formation of intramyelinic edema and hemorrhagic foci within the neural tissue. Notably, these neurological and muscular degradations were virtually absent in the ALE-supplemented group, which remained compatible with the usual histological appearance of healthy fish, further emphasizing the systemic protective potential of artichoke leaf extract against pyrethroid-induced tissue damage.

Discussion

The present study demonstrated that deltamethrin exposure significantly suppresses the growth performance of Nile tilapia (G3), as evidenced by marked reductions in weight gain and specific growth rate (SGR), alongside an increased feed conversion ratio (FCR). This growth retardation likely stems from the metabolic cost of detoxifying pyrethroids, diverting energy from growth toward survival mechanisms. These findings are strongly supported by the recent toxicological assessments of Eli et al. (2025) and Li et al. (2025), who reported that deltamethrin and related pyrethroids cause systemic metabolic disorders that directly impair the growth of aquatic organisms.

The dietary inclusion of 2% artichoke leaf extract (ALE) in the G4 group effectively mitigated the adverse impacts of deltamethrin, restoring growth parameters toward control levels. This improvement aligns with the findings of Shohreh et al. (2025) and Esen et al. (2025), who collectively demonstrated that artichoke extract significantly enhances growth performance, digestive enzyme activities, and nutrient assimilation in Nile tilapia. Specifically, Shohreh et al. (2025) observed that ALE promotes the expression of growth-related genes and boosts antioxidant capacity under environmental stress. Similarly, Esen et al. (2025) confirmed that the bioactive compounds in *Cynara scolymus* support metabolic efficiency and improve feed utilization rates, which explains the enhanced weight gain and optimized feed conversion ratio (FCR) observed in the current study. These findings suggest that the phenolic and flavonoid-rich profile of artichoke extract acts as a functional metabolic enhancer, effectively neutralizing the growth-inhibitory effects of aquatic pollutants. Furthermore, the efficacy of using natural dietary additives to counteract chemical-induced growth suppression in this species is reinforced by the recent works of Öz et al. (2025), who found that dietary *Nigella sativa* oil successfully reversed the growth-inhibiting effects of diazinon exposure.

Additionally, Öz et al. (2026) highlighted a broader physiological trend where natural plant-based supplements serve as potent metabolic stabilizers, protecting the systemic health of Nile tilapia against various aquatic pollutants such as boric acid. Consequently, the recovery observed in our ALE groups suggests that artichoke's rich phenolic content optimizes energy partitioning and promotes resilience, offering a sustainable strategy for maintaining fish productivity in contaminated aquaculture environments.

In the present study, significant decreases were observed in fundamental hematological parameters, including red blood cell count (RBC), hemoglobin (Hb), and hematocrit (HCT) in the G3 group exposed to deltamethrin. These results align with the hematological toxicity findings reported by David et al. (2015) and Gewaily et al. (2021), confirming that pyrethroids induce anemia by suppressing the hematopoietic system. Such deteriorations in erythrocyte indices inevitably lead to a reduction in oxygen-carrying capacity, potentially causing systemic hypoxia in fish tissues. This toxicological pattern is further corroborated by the recent work of Öz et al. (2025), who demonstrated that pesticide exposure leads to marked erythropoiesis inhibition in Nile tilapia.

In contrast, these parameters showed substantial recovery in the ALE-supplemented G4 group, with erythrocyte values approaching control levels. This improvement is consistent with the findings of Esen et al. (2025) and Shohreh et al. (2025), who reported that *Cynara scolymus* extract stabilizes red blood cell indices and enhances the physiological resilience of Nile tilapia under chemical and environmental stress. Furthermore, Öz et al. (2026) recently highlighted that natural antioxidant-rich supplements can counteract blood-related toxicity by protecting the cell membrane integrity of erythrocytes from oxidative damage. Therefore, the restorative capacity of ALE in our study can be attributed to its high concentration of phenolic compounds, which exert potent antioxidant and anti-inflammatory effects, thereby neutralizing the hematotoxic impacts of deltamethrin as similarly noted by Elsayyad et al. (2024).

The biochemical profile of fish is a vital indicator for monitoring physiological changes and organ health under toxic stress. In the present study, deltamethrin exposure (G3) led to a significant increase in serum glucose, cholesterol, and liver enzyme activities (AST, ALT, and ALP). This hyperglycemic and hyperlipidemic response can be attributed to the mobilization of energy reserves to cope with the increased energy demand caused by pesticide-induced stress. These findings are strongly supported by Kumari and Mishra (2025), who demonstrated that deltamethrin significantly disrupts enzymatic balances, particularly

affecting alkaline and acid phosphatases, leading to metabolic instability in fish. Furthermore, the elevation of AST and ALT levels in our study indicates hepatic cellular damage, a phenomenon recently confirmed by Li et al. (2025), who reported that deltamethrin triggers severe liver damage and metabolic disorders in Japanese flounder.

On the other hand, the administration of 2% artichoke leaf extract (ALE) in the G4 group significantly stabilized these biochemical parameters, bringing glucose and enzyme levels closer to the control values. This hepatoprotective and metabolic-regulatory effect of ALE is consistent with the recent findings of Rabea et al. (2025), who observed that natural organic acidifiers could mitigate deltamethrin-induced systemic disruptions in Nile tilapia. The restorative capacity of artichoke extract likely stems from its rich phenolic composition, which, as noted by Eroğlu et al. (2026) in similar natural antioxidant studies, helps maintain enzymatic homeostasis and protects organ integrity against pyrethroid toxicity. Consequently, these results suggest that ALE acts as a potent metabolic stabilizer, counteracting the enzyme leakage and metabolic shifts recently characterized in toxicological models by Eli et al. (2025).

In the oxidative stress parameters evaluated within the scope of the study, a significant increase in MDA levels and a substantial decrease in antioxidant enzyme activities, such as SOD, CAT, and GPx, were detected in the G3 group under deltamethrin exposure. These findings are consistent with previous reports which demonstrated that pesticides weaken antioxidant defense mechanisms by escalating the production of reactive oxygen species (ROS). More recently, Wu et al. (2025) reinforced this mechanism by showing that deltamethrin triggers systemic oxidative stress through the modulation of Nrf2-mediated pathways in fish. Similarly, Eroğlu et al. (2026) reported that deltamethrin exposure leads to a marked inhibition of essential antioxidant enzymes like paraoxonase and arylesterase.

However, a significant increase in antioxidant enzyme activities and a decrease in MDA levels were observed in the G4 group supplemented with artichoke leaf extract (ALE). This reveals that ALE creates a robust protective barrier for the antioxidant system and suppresses oxidative damage at the cellular level. This restorative effect is mirrored in recent studies where natural antioxidants were used to combat pyrethroid toxicity; for instance, Wu et al. (2025) demonstrated that antioxidant supplementation could reverse deltamethrin-induced oxidative damage, and Eroğlu et al. (2026) highlighted the efficacy of natural phenolic-rich additives in restoring oxidative balance. These results are further supported by Elsayyad et al. (2024), who reported that *Cynara scolymus* extract supports the antioxidant defense system against chemical-induced stressors. The synergy between the phenolic compounds in ALE and the fish's endogenous defense mechanisms likely provides the holistic protection observed in our study, effectively neutralizing the metabolic disruptions recently characterized by Kumari and Mishra (2025).

Interestingly, the total oxidant status (TOS) and oxidative stress index (OSI) in the G3 group (deltamethrin-exposed) were found to be lower than those in the control group. While this may initially appear contradictory, it is likely attributed to a potent compensatory response of the antioxidant defense system. As evidenced by our results, deltamethrin exposure induced a robust mobilization of endogenous antioxidant enzymes, specifically SOD and GPx. While acute exposure studies, such as those conducted by Amin and Hashem (2012) for 48 h and Ullah et al. (2019) for 96 h, typically report immediate increases in oxidant status and depletion of defenses, our 30-day sub-chronic findings suggest a transition to a hormetic phase. This massive enzymatic mobilization appears to have effectively sequestered and neutralized reactive oxygen species (ROS), reducing the measurable total oxidant load (TOS) below baseline levels at the final sampling point. Such over-compensation mechanisms, where the organism's scavenging capacity temporarily

exceeds the rate of oxidant production as a survival strategy, align with the metabolic distress patterns and subsequent adaptive measures described in Nile tilapia under prolonged pesticide stress (El-Sayed and Saad 2008).

Gills of fish perform vital functions such as respiration, osmoregulation, and acid–base balance. These complex structures provide effective gas exchange between water and fish. Thanks to their large surface areas, they adjust ion levels and enable fish to survive in changing environmental conditions (Santos et al. 2019). Pesticides, which are among the important pollutants of aquatic ecosystems, can cause various histopathological lesions and structural changes in the gill tissue, which are directly related to the impairment of respiratory function in aquatic organisms. One of the most frequently observed histopathological changes in fish gills exposed to pesticides is epithelium detachment. This change occurs as the separation of epithelial cells from the underlying tissue, leading to dysfunction in respiratory efficiency and increased tissue sensitivity (Velmurugan et al. 2009). Similarly, Okogwu et al. (2022) reported significant disorganization and fragmentation in gill structures in studies conducted on African catfish (*Clarias gariepinus*), and it was shown that exposure to agricultural pesticides disrupted the morphology of gill tissues and potentially exacerbated respiratory problems (Okogwu et al. 2022). However, the coexistence of specific histopathological findings such as hyperplasia of epithelial cells, necrosis and fusion of gill lamellae indicate that the respiratory surface area, which is critical for gas exchange, is negatively affected (Shah and Parveen 2022). This degeneration in the gill structure is directly related to respiratory dysfunction and can potentially lead to hypoxia and associated physiological stress (Öz et al. 2024a). Histopathological changes observed in the gill tissue of fish exposed to commonly used agricultural pesticides; These changes include findings such as epithelial detachment, hyperplasia, lamellae fusion and telangiectasia, all of which are significantly correlated with respiratory dysfunction. These changes reveal the multifaceted effects of aquatic pollutants on fish physiology and emphasize the need for meticulous environmental monitoring and regulatory measures regarding pesticide applications (Yancheva et al. 2019) (Figs. 1, 2, 3 and 4).

Understanding these dynamics ultimately supports the broad ecological impacts and conservation strategies required for the sustainability of aquatic life. Exposure of fish to agricultural pesticides leads to significant histopathological changes, especially in the liver, which is a major organ involved in detoxification processes. Various studies have demonstrated that distinct morphological changes such as hepatocyte hypertrophy and necrosis occur in liver tissue as a result of pesticide exposure. Pawar and Shrivastava suggest that changes in liver morphology serve as indicators of exposure to environmental stressors and may provide important information about the biochemical pathways affected by these toxic compounds (Pawar and Shrivastava 2023). Acute and chronic exposures to pesticides manifest themselves in the form of distinct histopathological patterns. Akeredolu and colleagues studied the differences in histopathological responses of *Clarias gariepinus* to both acute and sub-lethal pesticide exposures and reported varying degrees of liver damage depending on the intensity and duration of exposure (Akeredolu et al. 2022). Various types of pesticides exhibit different histopathological findings. For example, chlorpyrifos, an organophosphate, has been shown to cause severe structural changes in fish liver characterized by hepatocyte necrosis and bile duct proliferation, reflecting a cholestatic effect (Topal et al. 2014). Other pesticides such as neonicotinoids similarly cause liver architectural deterioration, leading to inflammatory responses and dysregulated hepatic function (Azadikhah et al. 2023). In cypermethrin toxicity, hemorrhage, vacuolar and hydropic degeneration in the liver sinusoids have been reported (Öz et al. 2024a, 2024b). Exposure of fish to common agricultural pesticides particularly affects histopathological structures in the brain,

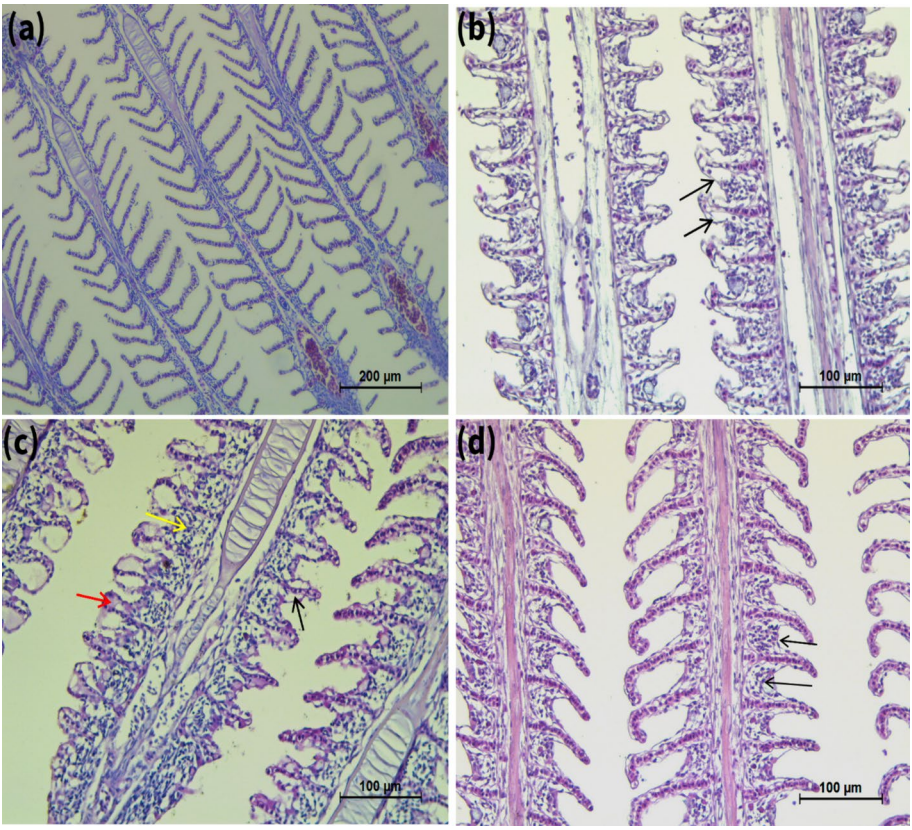


Fig. 1 Gill tissue of Nile tilapia (*Oreochromis niloticus*). (**A**) Normal histological image of gills of Nile tilapia (*O. niloticus*); Group 1, hematoxylin–eosin stain (H-E) (Scale bar = 200 µm). (**B**) Detachment of epithelial cells of secondary lamellae (black arrow); Group 2, hematoxylin–eosin stain (H-E) (Scale bar = 100 µm). (**C**) Edema and desquamation of secondary lamellae in the gills (red arrow), fusion of lamellae (yellow arrow), detachment of epithelial cells of secondary lamellae (black arrow), Group 3, hematoxylin–eosin stain (H-E) (Scale bar = 100 µm). (**D**) Fusion of secondary lamellae (black arrow), Group 4, hematoxylin–eosin stain (H-E) (Scale bar = 100 µm)

causing profound consequences on neurological functions and general health. Organophosphates, which are widely used as pesticides, exhibit neurotoxic effects by inhibiting cholinesterases and lead to significant neurochemical disorders. When the bioconcentration of methyl parathion was examined in various fish species, a significant accumulation of this pesticide in brain tissues at concentrations up to 80 ppm was observed within the first 30 min of exposure, affecting cholinergic neurotransmission, which is critical for the maintenance of neurological functions (Bosco de Salles et al. 2015).

Studies have identified significant histopathological changes in fish brains after pesticide exposure, including neuronal degeneration, vacuolization, and hyperemia (Yildirim et al. 2006; Amin et al. 2023). Morphological changes such as neuronal degeneration, necrosis, and synaptic changes have been associated with both chlorinated pesticides and organophosphates. These changes negatively affect behavioral outcomes, as impaired neuroplasticity and neuronal communication can lead to disorganized movements and cognitive dysfunction in fish (Doherty et al. 2016; Gandar et al. 2016). Similar neurological

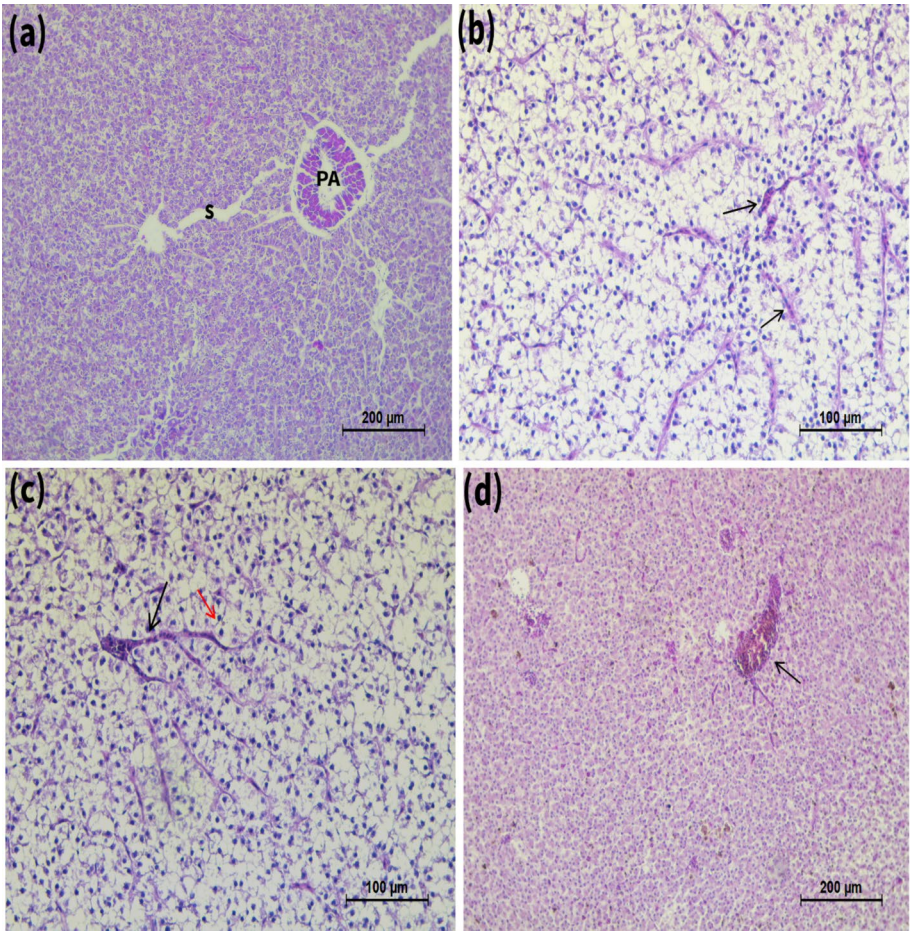


Fig. 2 Liver tissue of Nile tilapia (*Oreochromis niloticus*). (**A**) Histological images of normal liver of Nile tilapia (*O. niloticus*); exocrine pancreatic acini (PA) and liver sinusoid (S) Group 1, hematoxylin–eosin stain (H-E) (Scale bar = 200 µm). (**B**) Congestion in the liver (black arrow); Group 2, hematoxylin–eosin stain (H-E) (Scale bar = 100 µm). (**C**) Vacuolar degeneration and hydropic degeneration of hepatocytes (red arrow) in the liver, congestion in the liver (black arrow); Group 3, hematoxylin–eosin stain (H-E) (Scale bar = 100 µm). (**D**) Congestion in the liver sinusoids (black arrow); Group 4, hematoxylin–eosin stain (H-E) (Scale bar = 200 µm)

disorders can cause functional losses in the gills of fish, leading to reduced oxygen uptake and toxin elimination capacities (Pallavi 2020). Muscle atrophy is mostly observed under conditions of long-term toxic exposure or severe stress. Chronic exposure associated with pesticides can trigger catabolism of muscle proteins, which leads to atrophy of muscle tissue.

While the present study provides significant insights into the mitigative potential of artichoke leaf extract, it has certain limitations that should be acknowledged to guide future research. First, a standardized commercial artichoke leaf extract was utilized to ensure practical applicability for the aquaculture industry. Although the primary bioactive markers of such extracts are well-documented, we did not perform batch-specific chemical characterization,

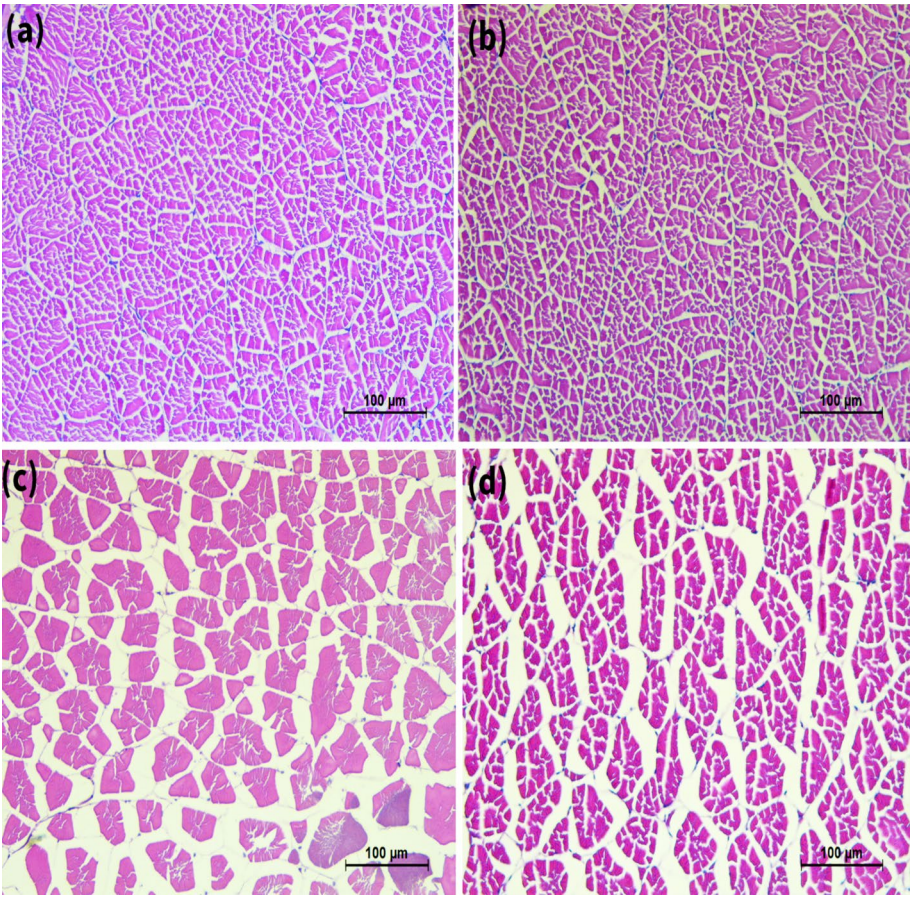


Fig. 3 Nile tilapia (*Oreochromis niloticus*) muscle tissue. (**A**) Normal histological image of Nile tilapia (*O. niloticus*) back muscle, Group 1 hematoxylin–eosin staining (H-E) (Scale bar = 100 µm). (**B**) No lesions were observed in the muscles, Group 2 (H-E) (Scale bar = 100 µm). (**C**) Severe muscle lesions. Atrophy image, Group 3 (H-E) (Scale bar = 100 µm). (**D**) Minimal muscle lesions, Group 4 (H-E) (Scale bar = 100 µm)

such as HPLC analysis, in our laboratory. Second, this research evaluated a single dietary inclusion level of 2.00% ALE. While this concentration proved effective in mitigating deltamethrin-induced toxicity under the current conditions, future studies incorporating a wider range of concentrations are necessary to establish a precise dose–response relationship. Additionally, the 30-day experimental duration primarily reflects sub-acute physiological responses. Investigating longer-term chronic exposure would be valuable to determine the sustained protective efficacy and potential cumulative effects of ALE in larger-scale production systems. Addressing these factors in future trials will further enhance the precision of phytotherapeutic applications in Nile tilapia culture.

The histopathological alterations observed in the gills, liver, and brain (G3) provide a structural basis for the systemic disruptions recorded in growth and biochemistry. Specifically, the severe epithelial desquamation and lamellar fusion in the gills directly correlate with reduced respiratory efficiency and the subsequent metabolic stress

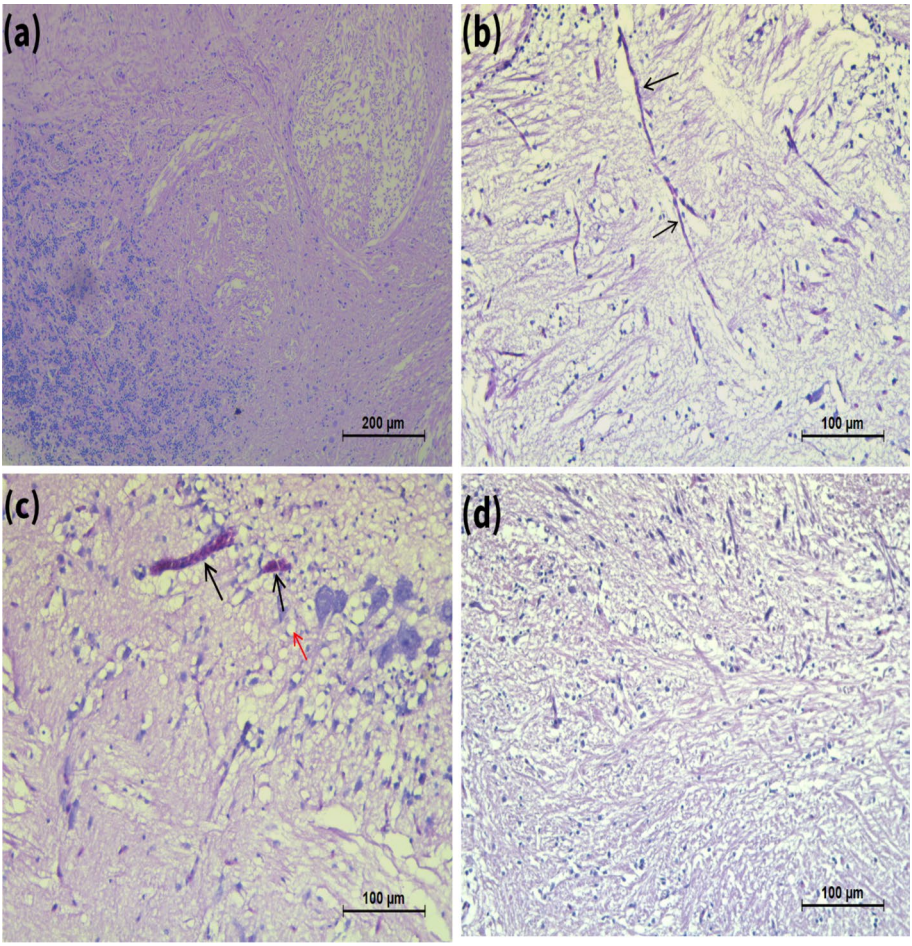


Fig. 4 Nile tilapia (*Oreochromis niloticus*) brain tissue. (**A**) Normal histological image of Nile tilapia (*O. niloticus*) brain, Group 1, hematoxylin–eosin stain (H-E) (Scale bar = 200 µm). (**B**) Hyperemia in the brain (black arrow), Group 2, hematoxylin–eosin stain (H-E) (Scale bar = 100 µm). (**C**) Vacuolization (red arrow) and hyperemia in the brain (black arrow), Group 3, hematoxylin–eosin stain (H-E) (Scale bar = 100 µm). (**D**) Histopathological image of Nile tilapia (*O. niloticus*) brain, Group 4, hematoxylin–eosin stain (H-E) (Scale bar = 100 µm)

reflected by hyperglycemic responses. In the liver, the extensive hydropic and vacuolar degeneration explains the leakage of hepatic enzymes such as AST, ALT, and ALP into the serum. Furthermore, the neuromuscular degradations, including intramyelinic edema and muscle atrophy, likely contributed to the significant growth retardation by impairing the fish's ability to forage and utilize nutrients efficiently. Conversely, the preservation of tissue architecture in the ALE-supplemented G4 group confirms that the biochemical and antioxidant improvements were rooted in stabilized cellular integrity, allowing for the complete recovery of growth performance even under toxicant exposure.

Conclusion

The present study demonstrates that deltamethrin exposure induces significant physiological disruptions in Nile tilapia, affecting growth performance, hemato-biochemical parameters, and tissue integrity. The observed alterations in hepatic enzyme activities, lipid peroxidation (MDA), and the shifts in oxidative stress markers (TAS, SOD, CAT, GPx, and TOS/OSI) reflect the multifaceted systemic stress caused by this pesticide.

Notably, the dietary inclusion of 2% artichoke (*Cynara scolymus*) leaf extract (ALE) showed potential in mitigating these adverse impacts. Fish supplemented with ALE exhibited improvements in growth parameters, partial restoration of hematological and biochemical indices, and enhanced antioxidant enzyme activities. These effects are likely associated with the bioactive phenolic and flavonoid constituents of the extract, which contribute to its hepatoprotective and antioxidant properties.

In conclusion, these findings suggest that artichoke leaf extract may serve as a beneficial functional feed additive to support fish health in aquaculture environments where pesticide exposure is a concern. However, further studies are recommended to determine the optimal dosage and evaluate the long-term efficacy of ALE under chronic exposure conditions.

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Author contributions Harun GÜMÜŞ: Investigation, Formal analysis, Data curation, review & editing Writing- Original draft preparation. Suat DIKEL: Methodology, review & editing Writing- Original draft preparation. Mustafa ÖZ: Conceptualization; Data curation; Formal analysis; Funding acquisition; Investigation; Methodology; Project administration; Resources; Software; Validation; Visualization; review & editing Writing- Original draft preparation.

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Data availability No datasets were generated or analysed during the current study.

Declarations

Ethics approval and consent to participate The experimental procedures of this study were conducted in strict accordance with ethical guidelines for animal research. The study protocol was reviewed and approved by the Çukurova University Aquatic Vertebrates Experiments Local Ethics Committee (Approval No: 4; Date: 07.05.2024). All efforts were made to minimize fish suffering and to reduce the number of animals used, in accordance with institutional and international animal welfare standards.

Consent for publication Not applicable.

Clinical trial number Not applicable.

Disclosure No potential conflict of interest was reported by the author(s).

Competing interests The authors declare no competing interests.

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