Environmental Pollution 348 (2024) 123853

Contents lists available at ScienceDirect

Environmental Pollution

journal homepage: www.elsevier.com/locate/envpol

Combined neurotoxicity of aged microplastics and thiamethoxam in the early developmental stages of zebrafish (*Danio rerio*)☆

Yanan Sun [a,b,1], Ping Ding [b,1], Jiayi Zhang [b,c], Kexin Sun [b], Xintong Li [a,b], Qing Ge [b,c], Yao Dang [b], Yunjiang Yu [b], Guocheng Hu [a,b,*]

[a] *The Postgraduate Training Base of Jinzhou Medical University (South China Institute of Environmental Sciences, Ministry of Ecology and Environment), Guangzhou, 510655, China*

[b] *State Environmental Protection Key Laboratory of Environmental Pollution Health Risk Assessment, South China Institute of Environmental Sciences, Ministry of Ecology and Environment, Guangzhou, 510655, China*

[c] *School of Public Health, China Medical University, Liaoning, 110122, China*

ARTICLE INFO

Keywords:
Microplastics
Thiamethoxam
Neurotoxicity
Photoaged
Zebrafish

ABSTRACT

Microplastics (MPs) pollution, together with its consequential effect on aquatic biota, represent a burgeoning environmental concern that has garnered significant scholarly attention. Thiamethoxam (TMX), a prevalently utilized neonicotinoid insecticide, is renowned for its neurotoxic impact and selective action against targeted pests. The aquatic environment serves as a receptacle for numerous pollutants, such as MPs and neonicotinoid insecticides. However, there is currently a lack of comprehensive understanding regarding the toxic effects of co-exposure to aged MPs and neonicotinoid insecticides in aquatic organisms. Therefore, we endeavor to elucidate the deleterious impacts of aged polystyrene (PS) and TMX on zebrafish (*Danio rerio*) larvae when present at environmentally relevant concentrations, and to reveal the underlying molecular mechanisms driving these effects. Our study showed that exposure to aged PS, TMX, or their combination notably inhibited the heart rate and locomotion of zebrafish larvae, with a pronounced effect observed under combined exposure. Aged PS and TMX were found to diminish the activity of antioxidative enzymes (SOD, CAT, and GST), elevate MDA levels, and disrupt neurotransmitter homeostasis (5-HT, GABA and ACh). Notably, the mixtures exhibited synergistic effects. Moreover, gene expression related to oxidative stress (e.g., *gstr1, gpx1a, sod1, cat1, p38a, ho-1,* and *nrf2b*) and neurotransmission (e.g., *ache, ChAT, gat1, gabra1, 5ht1b,* and *5ht1aa*) was significantly altered upon co-exposure to aged PS and TMX in larval zebrafish. In summary, our findings support the harmful effects of aged MPs and the neonicotinoid insecticides they carry on aquatic organisms. Results from this study enhance our understanding of the biological risks of MPs and insecticides, as well as help fill existing knowledge gaps on neonicotinoid insecticides and MPs coexistence toxicity in aquatic environment.

1. Introduction

Microplastics (MPs) represent a class of environmental contaminants characterized by their durability and hydrophobicity, which have become matters of increasing concern due to their omnipresence and proven toxicological effects on aquatic organisms (Khalid et al., 2021; Xu et al., 2020; Yu et al., 2022). Moreover, their propensity to adsorb organic pollutants from their surroundings has been particularly concerning, as this facilitates the transfer and potential bioaccumulation of pollutants within the environment (Torres et al., 2021; Wang et al., 2018; Wang et al., 2020a). Polystyrene (PS) is one of the predominant varieties of MPs found in oceans across the globe (Amelia et al., 2021). Its small size and buoyancy characteristics allow for easy ingestion by marine organisms across multiple trophic levels. Documented evidence illustrates its propensity for transfer and accumulation within the food web, resulting in biomagnification and consequent negative impacts on aquatic organisms (Assas et al., 2020; Huang et al., 2021; Wang et al., 2019).

☆ This paper has been recommended for acceptance by Maria Cristina Fossi.

* Corresponding author. The Postgraduate Training Base of Jinzhou Medical University (South China Institute of Environmental Sciences, Ministry of Ecology and Environment), Guangzhou, 510655, China.

E-mail address: huguocheng@scies.org (G. Hu).

[1] These authors contributed equally to this work.

https://doi.org/10.1016/j.envpol.2024.123853
Received 18 December 2023; Received in revised form 25 February 2024; Accepted 22 March 2024
Available online 27 March 2024
0269-7491/

Thiamethoxam (TMX), a neonicotinoid insecticide is known for its neurotoxic properties. Its rise in agricultural use can be attributed to its broad-spectrum effectiveness and potent neurotoxic impact on targeted pests (Gaboardi et al., 2023). As an emergent insecticide, TMX residues have been detected in the aquatic environments, occurs in the Venice Lagoon (Italy), the Seto Inland Sea (Japan), and various tributaries of the Pearl River, such as the Guangzhou reach (Hano et al., 2019; Pizzini et al., 2024; Xiong et al., 2019). Beyond its widespread distribution, exposure to TMX is linked to a host of deleterious impacts on aquatic organisms, ranging from oxidative stress to developmental and neurotoxic effects (Sun et al., 2022; Yang et al., 2022; Yang et al., 2023b). When MPs coexist with insecticides in the environment, the MPs may serve as carriers by adsorbing the insecticides onto their surfaces (Li et al., 2021; Wang et al., 2020b). This interaction could culminate in synergistic contamination that poses a threat to aquatic organisms and has the potential to disrupt entire aquatic ecosystems. For instance, the addition of polyvinyl chloride (PVC) particles was observed to exacerbate the toxicity of imidacloprid (IMI, another neonicotinoid insecticide) to the freshwater dipteran (*Chironomus riparius*) (Scherer et al., 2020; Luo et al., 2021). Another study revealed that co-exposure to PS and IMI, even at low concentrations, induced more significant hepatotoxicity in zebrafish (*Danio rerio*) than exposure to either alone. Similarly, chlorpyrifos-absorbed PS was found to have a more pronounced effect on zebrafish (Huang et al., 2023). These investigations frequently utilize virgin MPs as a standard model for study. However, MPs invariably undergo aging processes in natural environmental settings, including exposure to ultraviolet (UV) radiation, which may modify their toxicity characteristics (Chen et al., 2023a). The concurrent exposure of aquatic organisms to aged MPs and neonicotinoid insecticides may pose a risk, yet the toxicological effects remain to be fully elucidated. Therefore, further research is needed to clarify the combined toxic effects and underlying mechanisms of aged MPs loaded with neonicotinoid insecticides on aquatic organisms.

Zebrafish are widely regarded as excellent aquatic model organisms for investigating the harmful impacts of environmental chemicals. their short reproductive cycle, robust reproductive capacity, transparent embryos, a well-defined genetic background, and a high similarity to the human genome (Yuan et al., 2022). In this study, we focused on evaluating the individual and synergistic neurotoxic effects of aged PS and TMX at environmentally relevant concentrations on zebrafish larvae by examining the mechanisms through the detection of enzyme activities, neurotransmitter levels, and gene expression. The results provide insights into the neurotoxicity and potential risks associated with aged PS and neonicotinoid insecticides.

2. Materials and methods

2.1. Reagents and materials

Thiamethoxam (TMX, CAS No. 153719-23-4) with 99.70% purity were purchased from Dr. Ehrenstorfer GmbH Co., Ltd (Germany). Dimethyl sulfoxide (DMSO) used as cosolvent, was obtained from Aladdin Biochemical Technology Co., Ltd (Shanghai, China). Virgin PS (a size of 1 μm) samples were purchased from Janus New-Materials Co. (Nanjing, China).

Virgin PS samples were placed into quartz glass containers and subjected to continuous UV exposure at 26 °C for 40 days, using a UV lamp equipped with a 4 × 15 W bulbs that emitted at 254 nm. The aged PS samples were collected by rinsing them with ultrapure water and then heated at 60 °C to remove any residual contaminants. The physicochemical characterizations of virgin PS and aged PS can be found in Text S1.

2.2. Zebrafish cultivation and embryo obtainment

Adult wild-type zebrafish (AB strain, 4-month-old, female length: 3.14 ± 0.11 cm, mass: 0.44 ± 0.12 g; male length: 3.17 ± 0.13 cm, mass: 0.43 ± 0.11 g) were obtained from Nanjing EzeRinka Biotechnology Co., Ltd (Nanjing, China) and maintained according to the normative zebrafish protocols (Xiang et al., 2023). Prior to spawning, female and male zebrafish (at a 2:3 female/male ratio) were segregated using a transparent dam-board and housed in breeding tanks overnight. The following morning, the zebrafish were exposed to light for 30 min to stimulate spawning, after which they were paired for spawning by removing the dam-board. The procedure for collecting the embryos was performed in accordance with the method described by (Zhang et al., 2021).

2.3. Semi-static toxicity test

We selected 2 hpf embryos with normal development and randomly divided them among the blank control group and three treatment groups (aged PS, TMX and their mixture). Based on previous studies and the concentrations observed in the aquatic environment, we selected the concentrations of aged PS and TMX (1 μg/L and 1.5 μg/L, respectively) (Al-Sid-Cheikh et al., 2018; Das et al., 2023; Schaafsma et al., 2015). Each group consisted of three replicates with 100 embryos each. The embryos were hatched at 28.0 °C under a daily light and dark cycle (14 h:10 h) until reaching 120 hpf by semi-static toxicity test. After 120 hpf, larvae from each group were collected in 1.5 mL enzyme-free EP tubes (Axygen, US). Then immediately transferred to liquid nitrogen, and stored at - 80 °C for subsequent biochemical analysis and RT-qPCR.

2.4. Morphological analysis of zebrafish embryos and larvae

The embryos/larvae were checked under a stereomicroscope every 24 h for morphological changes and developmental abnormalities. Dead individuals were timely removed and counted. At 72 hpf and 96 hpf, Heart rates and hatchability of the larvae were counted, respectively. Abnormality was recorded at 120 hpf, and the body length (from the mouth to the end of the tail) of the larvae was measured at the same time.

2.5. Behavior assay of zebrafish larvae

At 120 hpf, larvae locomotor activity was evaluated using the ZebraLab Tracking system (ViewPoint Life Science, France). Initially, a test plate with 24 randomly chosen larvae for each treatment was placed in a quiet and dark environment for 40 min before testing. Then, two periods were recorded: the first in light and the second in darkness, simulating a light-dark transition, each lasting 10 min. This procedure was repeated twice for a total duration of 40 min. Raw swimming data were collected every 60 s, including the total duration of movements, total distance traveled, and then work out the swimming speed.

2.6. Biochemical analysis

After exposure, the levels of serotonin (5-HT), acetylcholine (ACh), gamma-aminobutyric acid (GABA), as well as the activities of catalase (CAT), superoxide dismutase (SOD), and glutathione-S-transferase (GST), and the content of malonaldehyde (MDA) in each treatment group were examined using ELISA kits obtained from Jiangsu Meimian industrial Co., Ltd., China. All indicators were measured following the manufacturer's instructions provided in Text S2.

2.7. Gene expression measurement

Total RNA was extracted using RNA extraction kit (TIANGEN, China), following the instructions provided by the manufacturer. The construction of the first-strand cDNA and RT-qPCR were performed according to the protocols provided by ACCURATE BIOLOGY, China. The *β-actin* gene was chosen as an internal control in the present study, and the primer sequences for the target genes are listed in Table S1. Finally, the relative gene expressions were determined using the $2^{-\Delta\Delta Ct}$ method.

2.8. Statistical analysis

In this study, statistics and correlation analyses of experimental data was conducted using IBM SPSS 22. Differences between blank control group and the treatments (aged PS, TMX, Mix) were evaluated by One-Way ANOVA with Tukey's post hoc test. All statisticsal analysis results are reported as the mean ± standard deviation (SD) based on triplicate experiments.

3. Results and discussion

3.1. Effects of aged PS and TMX on the early development of zebrafish larvae

Heart rate and body length constitute essential metrics for evaluating the early developmental progression of zebrafish larvae (Gonçalves Í et al., 2020; Torres-Ruiz et al., 2021). In our study, all experimental treatments demonstrated no significant impact on the hatchability and abnormality rates of embryos (Fig. 1A and B). Nevertheless, exposure to aged PS and TMX independently resulted in a notable reduction in heart rate, decreasing by 7.72% and 6.69%, respectively (Fig. 1C; $P < 0.05$). When comparing the individual exposures to aged PS or TMX, the combined exposure showed a markedly greater inhibitory effect on both heart rate (decreased by 1.65% and 2.73%, respectively) and body length (decreased by 3.17% and 2.84%, respectively; Fig. 1C and D; $P < 0.05$).

Statistical analysis of the data indicated that exposure to environmentally relevant concentrations of aged PS and TMX had no significant impact on the survival indices of zebrafish larvae, such as hatchability and abnormality rates ($P > 0.05$). However, aged PS and TMX negatively influenced growth-related parameters, particularly heart rate. These findings align with the observations made by (Prata et al., 2022), who reported that environmental concentrations of weathered PS diminished both heart rate and body length in zebrafish larvae, though without significantly affecting mortality, malformation, or hatching rate. Complementing this, (Erhunmwunse et al., 2023) demonstrated that exposure to 100 μg/L of IMI decreased heart rate in catfish (*Clarias gariepinus*) larvae.

Our study builds upon these insights, revealing a more pronounced toxic effect when aged PS is combined with TMX on zebrafish larval physiology. (Bhagat et al., 2021) observed that simultaneous exposure to PS and fluconazole, an azole fungicide, considerably inhibited the heart rate in zebrafish larvae, potentially due to increased reactive oxygen species production. Recently study showed that a binary mixture of PS and polybrominated diphenyl ethers (PBDEs) significantly escalated mortality and diminished heart rates in zebrafish larvae, likely related to an intensified oxidative stress response from combined exposures (Wang et al., 2023). Taken together, these studies suggest that marked developmental perturbations observed in co-treated groups could be ascribed to intensified oxidative stress responses induced by the combination of MPs with environmental contaminants. This hypothesis may elucidate observed reductions in heart rate and body length in zebrafish larvae subjected to aged PS and TMX in our research, meriting further exploration. Thus, our findings highlight synergistic toxicity from the co-exposure to aged PS and TMX, exerting detrimental effects on embryonic development.

3.2. Effects of aged PS and TMX on the locomotor activity of zebrafish larvae

The light/dark locomotor response serves as a sensitive, visually observable indicator of potential neurotoxicity in zebrafish exposed to target compounds (Chen et al., 2021b). Fig. 2A illustrates the assessment of swimming competence in zebrafish larvae subjected to aged PS, TMX, and their mixture (Mix) under alternating light and dark conditions. Compared to the control, a significant reduction in swimming speed was detected in larvae exposed to aged PS and TMX, both individually and in combination ($P < 0.05$; Fig. 2B). Specifically, the average swimming velocity was reduced by 29.13% for aged PS, 31.40% for TMX, and 45.13% for the Mix. Notably, the Mix group exhibited a pronounced

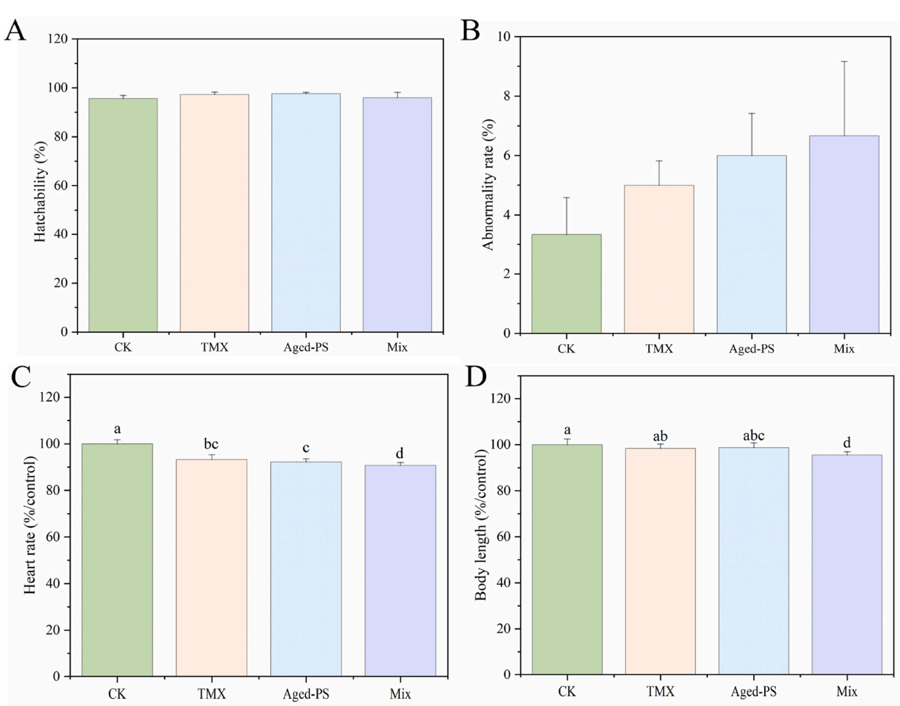

Fig. 1. Development and growth indices of zebrafish embryo and larvae in each treatment group. (A) Hatching rate. (B) Abnormality rate. (C) Heart rate. (D) Body length. The bars sharing different lowercase letters indicate statistically significant differences among the respective groups ($P < 0.05$).

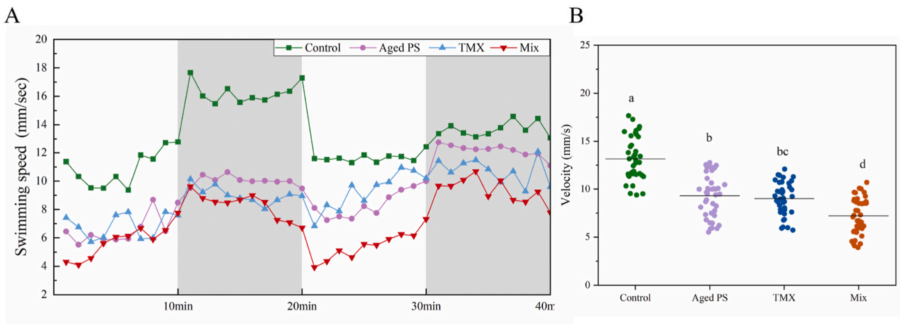

Fig. 2. Alterations in the locomotor activity of zebrafish larvae at 120 hpf. (A) Alterations in locomotor behavior traces observed during the dark/light photoperiod stimulus examination conducted at 120 hpf. (B) The velocity of 120 hpf zebrafish larvae during the light/dark photoperiod stimulus. The bars sharing different lowercase letters indicate statistically significant differences among the respective groups (*P < 0.05*).

decrease in swimming speed compared to individual exposures to aged PS or TMX, with reductions of 22.58% and 20.01%, respectively (*P < 0.05*). These findings indicate an elevated impact on locomotor activity from the combined exposure relative to either aged PS or TMX alone.

In this study, we noted a synergistic reduction in the swimming velocity of zebrafish larvae upon co-exposure to aged PS and TMX. Correspondingly, previous studies have reported that individual exposure to aged PS and TMX attenuates the locomotory activity of zebrafish (Hawkey et al., 2023; Zhang et al., 2021). Moreover, the combined exposure to MPs and other environmental pollutants has also been shown to substantially suppress the locomotion metrics of aquatic organisms. For example, co-exposure to PS and certain insecticides has been linked to diminished zebrafish larval locomotion, impacting parameters such as distance, velocity, and stasis (Varshney et al., 2023). The composite presence of PS and glyphosate has been associated with a reduction in the swimming distance of *Daphnia magna* (Nogueira et al., 2022), while another investigation found that the joint exposure to PS and penicillin (antibiotics) synergistically curtailed spontaneous movement and swimming behavior in zebrafish embryos (Chen et al., 2023b). Hence, it is crucial to embark upon additional research endeavors to acquire a more profound comprehension of the toxicity mechanisms underlying the observed alterations in locomotor behavior within this study.

3.3. Effects of aged PS and TMX on the oxidative stress of zebrafish larvae

To elucidate the potential neurotoxic mechanisms elicited by aged PS, TMX, and Mix, we assessed the activity of antioxidant enzymes (CAT, SOD), the detoxification enzyme (GST), and the content of lipid peroxides (MDA) in zebrafish larvae. Across all treated groups, a significant reduction in the activities of SOD, CAT, and GST was observed (Fig. 3A-C; *P < 0.05*), concomitant with a notable elevation in MDA levels (Fig. 3D; *P < 0.05*) compared with the control. Particularly, the co-exposure group manifested a heightened MDA content (3.73 ± 0.06 nmol/mg) when contrasted with the groups exposed to aged PS or TMX alone (2.69 ± 0.05 and 2.27 ± 0.04 nmol/mg, respectively), suggesting that oxidative damage was more pronounced under combined exposure conditions.

SOD, CAT, and GST are integral components of the antioxidant defense mechanism, functioning to inhibit, metabolize, or eliminate oxidatively compromised molecules (Chen et al., 2021a; Umamaheswari et al., 2021). MDA, the terminal product of lipid peroxidation in cell membranes, is widely recognized as an indicator of oxidative stress

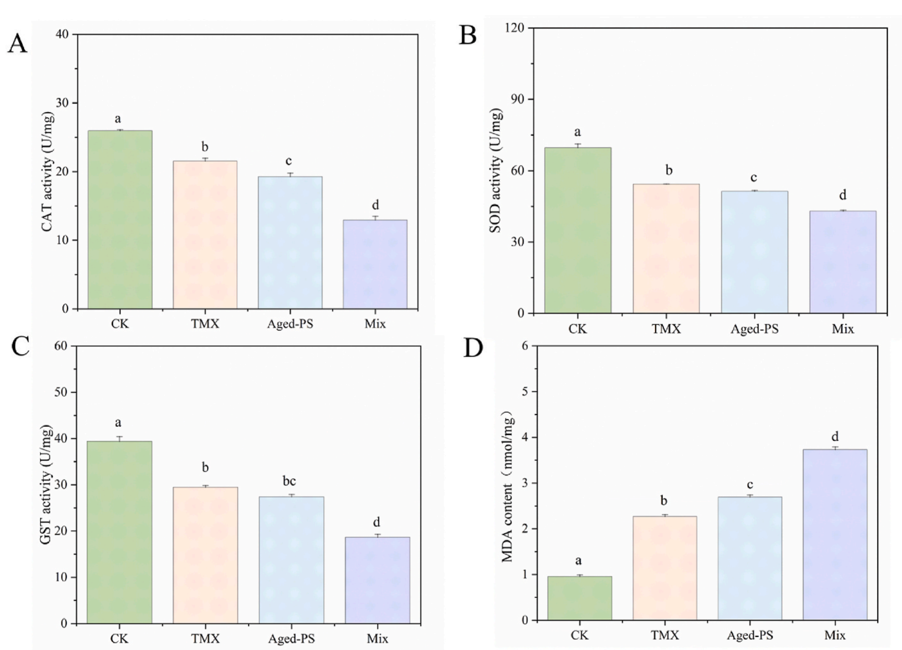

Fig. 3. Oxidative stress-related parameters in zebrafish larvae. The activity of (A) CAT, (B) SOD, (C) GST, and the content of (D) MDA. The bars sharing different lowercase letters indicate statistically significant differences among the respective groups (*P < 0.05*).

(Rios-Fuster et al., 2021). In previous studies, these biochemical markers (SOD, CAT, GST, and MDA) have been routinely utilized to monitor the oxidative stress response in aquatic organisms subjected to environmental pollutants (Suman et al., 2021). For instance, exposure of carp (*Cyprinus carpio*) to PS induced oxidative stress, evidenced by a substantial decrease in the activities of SOD and CAT (Cui et al., 2023). Similarly, exposure of the freshwater fish *Catla catla* to TMX resulted in diminished activities of SOD, CAT, and GST in their gills (Veedu et al., 2022). In our study, exposure to either aged PS or TMX alone induced oxidative stress in zebrafish, as indicated by the elevated MDA levels. Corresponding findings have been reported where exposure to MPs or TMX led to a significant increase in MDA levels in fish species (Das et al., 2023; Kayis et al., 2019; Yedier et al., 2023), suggesting that exposure to MPs and TMX may induce oxidative stress responses in aquatic organisms.

The antioxidant defense system is integral to counteracting oxidative stress, primarily through the function of key enzymes including SOD, CAT, and GST. Nonetheless, the concurrent decreases observed in the activities of these enzymes, alongside elevated MDA levels, points to a compromised integrity of this antioxidant defense system. (Li et al., 2022) explored the joint toxicological impacts of MPs and polybrominated diphenyl ethers (PBDEs) on the grouper (*Epinephelus moara*), identifying a decrease in SOD and GST activities accompanied by a marked rise in MDA levels in groups subjected to both contaminants. Similarly, (Aliakbarzadeh et al., 2023) reported that a binary mixture of PS and nonylphenol diminished CAT activity and glutathione (GSH) levels, which in turn induced oxidative stress and neurotoxicity in zebrafish. Our present study aligns with these findings, showing that co-exposure to aged PS and TMX led to a notable reduction in SOD, CAT, and GST activities in zebrafish larvae, alongside a significant elevation in MDA content. Considering the data presented, it is plausible to conjecture that the co-exposure to aged MPs and TMX could exacerbate oxidative stress through a synergistic inhibition of the antioxidant defense system.

3.4. Effects of aged PS and TMX on the neurotransmitter contents of zebrafish larvae

To investigate the mechanisms underlying the locomotor behavioral abnormalities in zebrafish larvae, we quantified the alterations in neurotransmitter levels following exposure to aged PS, TMX, and their combination. Relative to the control group (4.42 ± 0.23 ng/mg), the content of ACh was significantly increased in response to aged PS and TMX (6.96 ± 0.16 and 5.12 ± 0.08 ng/mg, respectively; Fig. 4B; $P < 0.05$). Furthermore, the content of GABA was elevated in the larvae treated with aged PS(6.56 ± 0.30 μmol/L) compared to controls (4.13 ± 0.14 μmol/L; Fig. 4C; $P < 0.05$). The content of 5-HT exhibited a significant decrease in the aged PS or TMX treatment groups (91.87 ± 0.61 and 104.77 ± 2.41 ng/mg, respectively; Fig. 4A; $P < 0.05$) versus the control (124.22 ± 2.48 ng/mg). The alterations in the levels of ACh, GABA, and 5-HT were further amplified in the Mix group, with significant elevations observed (8.96 ± 0.11 ng/mg for ACh, 7.55 ± 0.20 μmol/L for GABA, and 79.35 ± 1.75 ng/mg for 5-HT) relative to exposures to TMX or aged PS alone. This suggests the potential for a synergistic interaction between aged PS and TMX, further perturbing the equilibrium of neurotransmitters.

During critical periods of central nervous system (CNS) maturation in juvenile fish, neurotransmitters such as 5-HT, ACh, and GABA are crucial, playing central roles in processes that range from motor function and circadian rhythm regulation (Horzmann and Freeman, 2016; Rico et al., 2011). 5-HT is implicated in motor control and diurnal rhythms. ACh is essential for neuromuscular junctions and modulates the release of other neurotransmitters, while GABA serves as the primary inhibitory neurotransmitter within the CNS. In a previous study, exposure to insecticides can disrupt the synthesis and expression of neurotransmitters, resulting in neurological deficits and atypical behaviors (Tufi et al., 2016). Recently, (Yang et al., 2023a) found that TMX could adversely impact the swimming behavior of adult zebrafish by disturbing neurotransmitter equilibrium. Additionally, zebrafish embryos exposed to PS have shown marked changes in neurotransmitter levels and locomotor activity (Jeong et al., 2022; Suman et al., 2023). These outcomes align with our observations, indicating that exposure to PS or TMX is associated with changes in neurotransmitter levels and resulting neurotoxicity in zebrafish. Moreover, our study highlights that concurrent exposure to aged PS and TMX results in more pronounced disturbances within the neurotransmitter systems, potentially underpinning the observed reduction in swimming velocity among zebrafish in the mixed-exposure group.

3.5. Effects of aged PS and TMX on the gene expression of zebrafish larvae

To further elucidate underlying mechanism of the neurological neurotransmitter homeostasis and oxidative stress disruption in larvae after co-exposure to aged PS and TMX, we conducted an analysis of mRNA expression levels for genes associated with oxidative stress (*gstr1, gpx1a, sod1, cat1, p38a, ERK3, p38b, ho-1, nrf2a,* and *nrf2b*) and neurotransmitters of 5-HT *(5ht1b, 5ht1aa, 5ht2c)*, ACh *(ache, chrnα7, ChAT)*, and GABA *(gat1, gabra1, gad1b)*. As shown in Fig. 5, there were notable alterations in the expression of genes implicated in oxidative stress (*gstr1, sod1, cat1, p38a, gpx1a, ho-1,* and *nrf2b*) and neurotransmitter-related genes (*ache, ChAT, gat1, gabra1, 5ht1b,* and *5ht1aa*) within the Mix group ($P < 0.05$).

Our study shows that simultaneous exposure to aged MPs and TMX significantly alters mRNA levels related to neurotransmission and oxidative stress. A previous study found that zebrafish subjected to PS

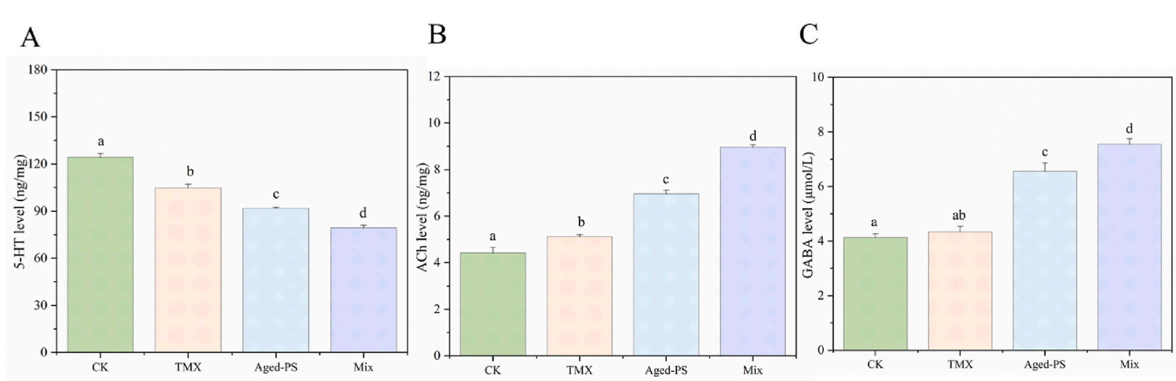

Fig. 4. The levels of neurotransmitters in zebrafish larvae. (A) 5-HT, (B) ACh, (C) GABA. The bars that share different lowercase letters indicate statistically significant differences among the respective groups ($P < 0.05$).

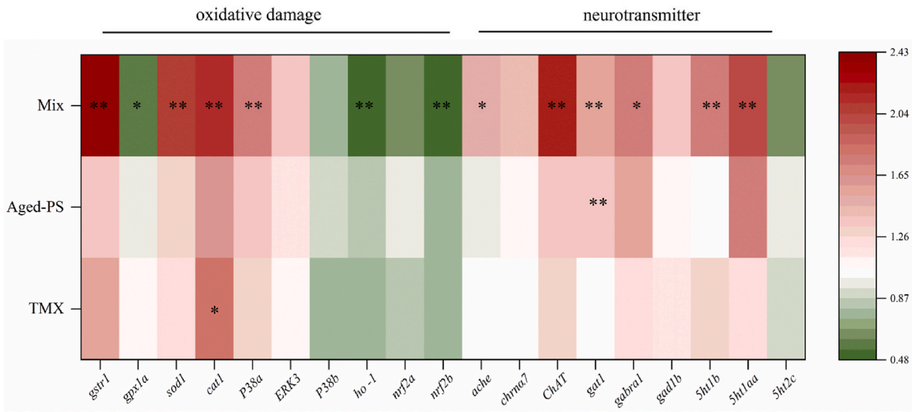

Fig. 5. The gene expression levels related to oxidative stress and neurotransmitters in zebrafish larvae. The statistical significance, when compared to the control group, was defined as **P < 0.05, **P < 0.01*.

exposure over 35 days exhibited an oxidative stress response, characterized by diminished gene expression levels of *cat*, *sod1*, *gpx1a*, and *ache* (Umamaheswari et al., 2021). Similarly, photo-aged PS was shown to interfere with the expression of oxidative stress-related genes (*p38a*, *cat1*, *sod1*, *gpx1a*, and *gstr1*) and neurotransmitter-related genes (*gat1*, *gabra1*, and *ache*) in zebrafish larvae (Ding et al., 2023). Other studies have documented the neurotoxic impacts of neonicotinoids on zebrafish, manifested through altered neurotransmitter gene expression (*gabra1*, *gad1b*, *ache*, and *gat1*), findings that align with our research (Xie et al., 2022; Zhang et al., 2021). Thus, oxidative damage and abnormal neurotransmission may be key mechanisms underlying the adverse effects caused by the coexistence of neonicotinoid insecticides and aged MPs in zebrafish larvae.

4. Conclusion

In conclusion, our findings demonstrate that zebrafish larvae exposed to environmentally relevant concentrations of aged PS and TMX exhibit significant growth inhibition and hypolocomotion. The disturbances in neurotransmission and oxidative stress identified are believed to play a pivotal role in the observed synergistic toxicity. Our research focused on the neuro effects of aged PS and neonicotinoid insecticides in fish. It is recommend that further studies be conducted on the potential effects of coexisting MPs with other organic pollutants to comprehensively understand the ecological risks associated with widespread MPs contamination.

CRediT authorship contribution statement

Yanan Sun: Writing – original draft, Investigation. **Ping Ding:** Writing – review & editing, Resources, Formal analysis. **Jiayi Zhang:** Methodology, Investigation. **Kexin Sun:** Writing – review & editing, Conceptualization. **Xintong Li:** Visualization, Investigation. **Qing Ge:** Investigation. **Yao Dang:** Writing – review & editing. **Yunjiang Yu:** Writing – review & editing, Supervision. **Guocheng Hu:** Resources, Data curation.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The authors are unable or have chosen not to specify which data has been used.

Acknowledgements

Financial support was received from the National Key R&D Program of China (2022YFC3902102).

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2024.123853.

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