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Pesticides and metal contaminants in Lake Chapala, Mexico: Differences in bioaccumulation and associated health risks

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ABSTRACT

Lake Chapala, Mexico's largest freshwater lake and primary drinking water source for approximately 5 million people, lacks comprehensive pesticide and metal contamination assessment across compartments. Unlike previous single-matrix or single-class studies, this is the first seasonal multi-matrix assessment of 17 pesticides and 8 heavy metals in water, sediment, and fish across eight stations over four hydroclimatic periods (2024-2025). The study targets the Poncitlán CKD hotspot and compares bioaccumulation patterns between native (*Ictalurus dugesii*) and invasive (*Oreochromis aureus, Cyprinus carpio, Micropterus salmoides*) fish species. Pesticide concentrations peaked at rainy season onset, with diuron (2824 ng/L), atrazine (1649 ng/L), and DDT/DDD detected at all sites. Glyphosate peaked during rain cessation (234,358.62 ng/L), suggesting erosion-dependent transport. These first multi-matrix seasonal patterns show that rainfall timing drives chemical mobilization and risk. Cadmium and lead exceeded Mexican regulatory limits in interstitial water at the Santiago-Zula confluence. Benthic-feeding fish accumulated significantly higher zinc (26.6 mg/kg) than pelagic species, irrespective of invasion status. A statistical correlation was observed between glyphosate and zinc in fish tissue, suggesting a possible co-occurrence pattern warranting further mechanistic validation. Ecological risk (ΣRQ) peaked during the rainy season (ΣRQ = 935.9), driven by DDD and diuron, with persistent year-round risk (ΣRQ 3.2-10.2). Although fish consumption hazard quotients were <1, multi-pathway community burden requires future study. These findings provide the first integrated baseline for this CKD-affected region and underscore the need for seasonal, multi-matrix surveillance that includes invasive species and co-contaminant interactions.

1. Introduction

Freshwater ecosystems are increasingly threatened by pollution, biological invasions, and habitat degradation (Reid et al., 2019; Tickner et al., 2020). Key contaminants, heavy metals, pesticides, and untreated wastewater, are being introduced at an unsustainable rate (Zhao et al., 2025), posing a major One Health risk with implications for ecological, biological, and human wellbeing (Domingo and Nadal, 2026).

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Water quality assessment relies on pollution indicators, including physicochemical parameters and contaminant concentrations, to evaluate ecosystem health and identify pollution sources (Mihali and Dippong, 2023; Dippong and Resz, 2022). The fate of these contaminants is compound-specific and governed by physicochemical and chemical structure-related fate parameters; chemicals with log $K_{ow} > 5$ and MW < 800 g/mol are likely to permeate biological tissues (Mackay et al., 2018; Miyata et al., 2022). Contaminants that exhibit bioaccumulative tendencies pose risks to human health via the consumption of contaminated produce (Rose, 2023).

Aquatic ecosystems and their food webs are inherently linked to human communities thus risk assessments need to account for the ecological interactions within the biota alongside broad spectrum pollutant analysis. Due to research constraints most studies focus on a single matrix, a single season, or a single contaminant class, leaving major gaps in risk evaluation (Ceschin et al., 2021). For instance, addressing only heavy metals or pesticides (Dippong et al., 2023, 2025; Dippong and Resz, 2024; Fures et al., 2026; Musa et al., 2025a; Leal-Acosta et al., 2022), which overlooks co-occurring pollutants. Moreover, most assessments are further limited to only one or two environmental matrices, such as water or sediment, rarely integrating biota. When biota are considered, studies typically focus on a single species (Musa et al., 2025b; Afzal et al., 2024; Akoto et al., 2016), failing to account for variations in feeding habits or ecological niches, which reduces the representativeness of the findings. This fragmented approach undermines a comprehensive understanding of real-world exposure and cumulative risks.

Biological interactions, including their direction and magnitude, are disrupted by chemical pollution in unpredictable ways, depending on taxon, chemical type, and concentration (South et al., 2019; Raman et al., 2024). These interactions may be further influenced by invasive species, which alter and reconfigure native species' interspecific interactions (South et al., 2025). When humans are incorporated into the food web, aquatic food consumption varies across demographics, potentially modulating the extent of exposure to harmful chemicals (South et al., 2025). A trait-based approach is needed, as benthic species tend to accumulate more sediment-bound contaminants than pelagic species (Leppänen, 1995; D'Avignon et al., 2023). However, to date, no field-based study has simultaneously integrated feeding habits, invasion status, and co-contaminant interactions (pesticides and heavy metals) within a single, seasonally resolved assessment of a large tropical lake system. This narrow focus constitutes a critical knowledge gap, especially in regions where pollution and biological invasions co-occur and directly affect human health.

Lake Chapala serves as Mexico's largest lake, providing more than 60% of Guadalajara's drinking water, sustaining local fisheries and biodiversity, and functioning as a tourist destination (SIAPA, 2024). Decades of unregulated discharge, including sewage, industrial, and agricultural runoff, have significantly degraded its water quality. As a result, the lake, which acts as the hydrologic terminus of the basin, has progressively accumulated persistent chemical pollutants. Notably, the northeast shoreline of Lake Chapala exhibits a high prevalence of chronic kidney disease (CKD) among local communities in the Poncitlán Municipality. While the epidemiology likely reflects a multifactorial etiology, local health officials and researchers have hypothesized a potential association with environmental exposures (García-García et al., 2020; Polanco-Flores, 2019; Sierra-Diaz et al., 2019), with pesticides and metals having been detected in residents' blood and urine (Sierra-Diaz et al., 2019; Lozano-Kasten et al., 2021). However, our study does not directly test this association; rather, we provide environmental occurrence data that may contribute to future exposure assessments.

Previous risk assessments conducted in Lake Chapala have been limited in scope thus we present the first comprehensive assessment of a broad spectrum of contaminants across multiple environmental matrices (water, sediment and biota), throughout a complete annual hydrological cycle. We expect that hydrological variation will alter contaminant concentration with a peak post-rainy season due to remobilization of sediment-bound pollutants. We used a trait-based approach to test whether benthic feeding habits or invasion status influences higher contaminant loads in native (*Ictalurus dugesii*) and invasive (*Oreochromis aureus, Cyprinus carpio, Micropterus salmoides*) fish species. We anticipated that invasive benthic fish would have a higher total contaminant load and investigated whether consumption of each species posed a significant human health risk.

Known contaminants in Lake Chapala include pesticides (DDT (dichlorodiphenyltrichloroethane), endosulfan, glyphosate, malathion), heavy metals (Al, As, Cr, Pb, Cd), and fecal coliforms (Gonzalez-Ocampo et al., 2022; de Anda et al., 2024;Ramírez-Sánchez and Fajardo-Montiel, 2024). Nevertheless, it is highly probable that other contaminants of emerging concern also exist.

Malathion concentrations in 2019 levels exceeded US EPA (Environmental Protection Agency) safety limits in 2019 (Silva-Madera et al., 2021). Regarding heavy metals, elevated concentrations of Al, As, Cr, Pb, and Cd in Lake Chapala exceed international guidelines (Ramírez-Sánchez and Fajardo-Montiel, 2024). However, a transport modelling study reported that most metals were below detection limits, attributing this discrepancy to variations in sampling timing and methods (Alvarez-Bobadilla et al., 2023). Despite these conflicting findings, discrepancies, bioaccumulation of Al, Sr, and Ba was confirmed in fish exposed to water and sediments, although concentrations remained below acute toxicity thresholds for wildlife (Torres et al., 2016).

Critically, previous investigations have been highly fragmented, as they typically assessed either pesticides or metals in isolation, seldom including all three matrices (water, sediment, biota), fail to cover all four hydroclimatic periods, and omit comparisons between native and invasive fish species. As a result, seasonal co-contaminant dynamics, the role of invasive species in bioaccumulation, and pesticide-metal synergies remain largely unexplored in the CKD-affected region of Lake Chapala. To contextualize the novelty of our approach, we compared the present study with previous investigations of pesticide and/or heavy metal contamination in Lake Chapala (Table S1). This comparison reveals that, unlike earlier studies, our work integrates both contaminant classes across all three matrices (water, sediment, biota) over a complete annual hydrological cycle, while explicitly accounting for feeding guild and invasion status, and evaluating co-contaminant interactions alongside ecological and human health risk.

Therefore, the primary research question guiding this study is how seasonal hydrology and fish ecological traits, specifically feeding habits and invasion status, influence the bioaccumulation of a broad spectrum of pesticides and heavy metals across water, sediment, and biota in a large, polluted tropical lake.

We hypothesize that seasonal variation in water flow and agricultural runoff will significantly affect contaminant concentrations, with peak levels expected during the post-rainy season owing to the remobilization of sediment-bound pollutants. Furthermore, we predict that invasive species will accumulate significantly higher total contaminant loads than native species, thus serving as major biovectors of sediment-bound pollutants. Finally, we expect that the co-occurrence of pesticides and heavy metals will yield measurable cumulative risks to both aquatic wildlife and local human consumers.

To test these hypotheses, we conduct the first comprehensive assessment of a broad spectrum of pesticides (both current-use and legacy compounds) and heavy metals across complementary environmental matrices, namely water, sediment and biota, throughout a complete annual hydrological cycle. Our study uniquely integrates ecological interactions by quantifying differences in bioaccumulation between native (*Ictalurus dugesii*) and invasive (*Oreochromis aureus, Cyprinus carpio, Micropterus salmoides*) fish species, explicitly examining whether benthic feeding habits or invasion status influences higher contaminant loads.

We seek to enhance our assessment of pollution in Lake Chapala by comparing bioaccumulation patterns between native and invasive fish species and by conducting a preliminary ecological and human health risk assessment. Two major pollutant classes, pesticides and metals, are of particular interest due to their no biodegradable characteristics and bioaccumulation potential, which pose significant risks to the ecosystem and human health (Ramírez-Sánchez and Fajardo-Montiel, 2024; Torres et al., 2016). These contaminants are introduced through anthropogenic activities such as industrial discharges, mining, and urban/agricultural runoffs.

Given these inputs, such pollutants are likely transported to Lake Chapala via the major inflow rivers (Lerma, Santiago, and Zula) and through diffuse runoffs from the Alto Lerma Watershed. Consequently, we anticipate higher concentrations near these sites. By integrating seasonal concentration data with ecological and human health risk quotients, this study provides the first integrated baseline for this system while identifying critical data gaps for future monitoring and policy, particularly the necessity of accounting for seasonal hydrology, invasive species, and co-contaminant interactions within water quality frameworks.

2. Materials and methods

2.1. Study area and sampling design

Lake Chapala is a shallow tropical lake (4-6 m deep) situated in west-central Mexico, spanning the border between the states of Jalisco and Michoacán. It is situated at an elevation of approximately 1515 m above sea level. Historically, the lake's surface area has varied between approximately 1100 and 1500 km^2, though in recent years it has averaged around 900 km^2 due to water abstraction and climate variability (Ramírez-Sánchez and Fajardo-Montiel, 2024; Alvarez-Bobadilla et al., 2023).

Lake Chapala's hydrology is characterized by a distinct wet-dry tropical climate: approximately 80% of the annual ~800 mm precipitation occurs during the rainy season (June–October), resulting in substantial inflows, whereas the remainder of the year is marked by dry conditions with gradually declining water levels (Greene, 2025). The principal inflow is the Lerma River, and the principal outflow is the Santiago River (Alvarez-Bobadilla et al., 2023).

The study focused on the northeastern portion of Lake Chapala near Poncitlán, an area influenced by the Lerma River and local agricultural tributaries, while also including sites located outside this region in order to capture broader spatial dynamics. Eight sampling stations (CH1-CH8) were selected based on Jalisco State Water Commission monitoring points, which represent river inflows, shoreline settlements, and open water (Fig. 1; Table S2).

Sampling campaigns were conducted in four periods: (i) rain onset (July 2024); (ii) rain cessation (late October 2024); (iii) cold-dry (February 2025); and (iv) warm-dry season (May 2025). On each sampling day, all sites were visited between 06:00h and 14:00 to ensure consistency and reduce diurnal variability in physicochemical parameters. During each campaign, samples were collected from all stations for water (surface and interstitial), sediment, and fish, resulting in 64 water

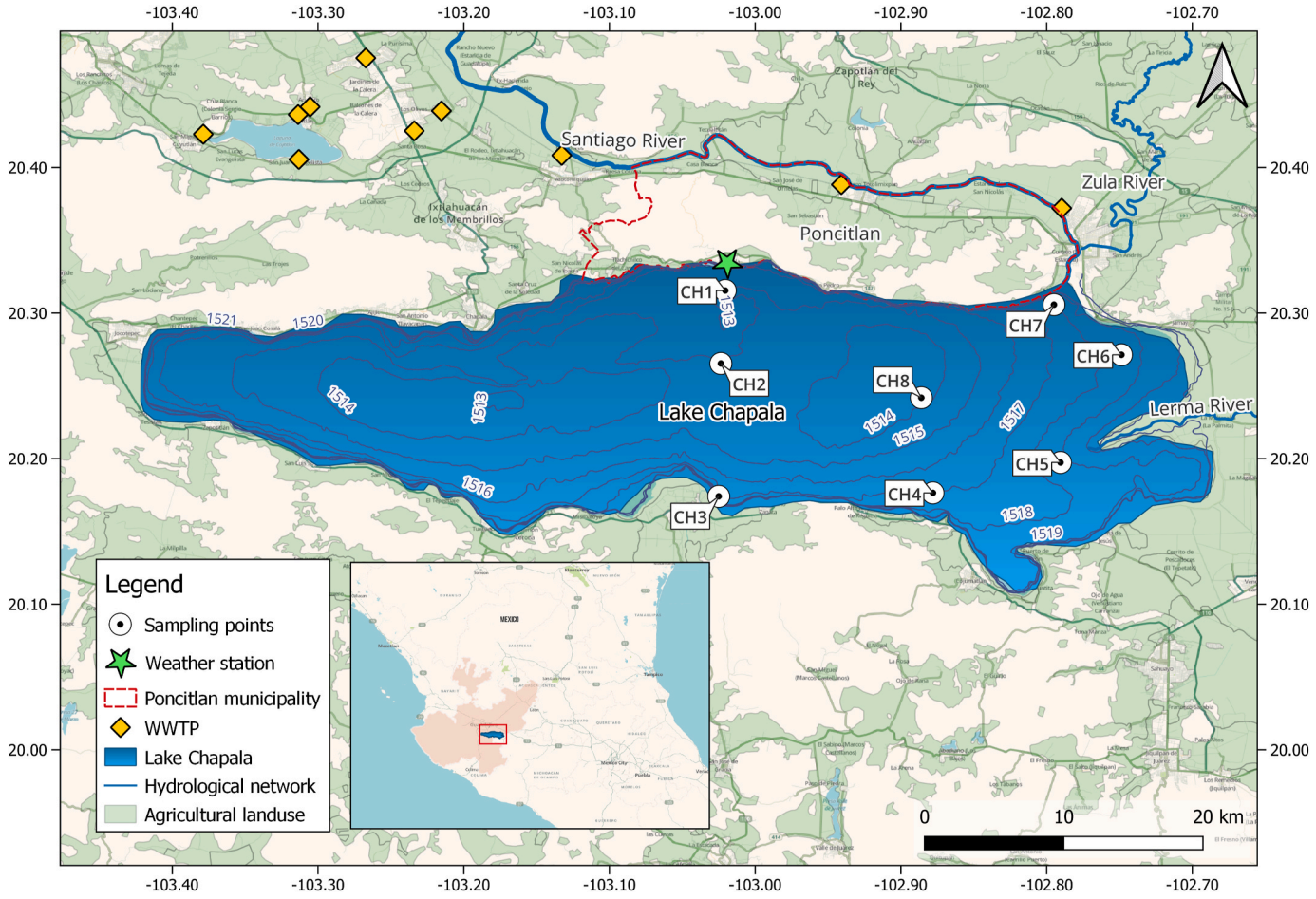


Fig. 1. Study area, sampling points and special features in Lake Chapala. WWTP: Wastewater treatment plant; Sampling points: CH1 to CH8. Coordinates: CH1 (20.31895°N, 103.04676°W); CH2 (20.27480°N, 103.04955°W); CH3 (20.19529°N, 103.03994°W); CH4 (20.17390°N, 102.87010°W); CH5 (20.21980°N, 102.78117°W); CH6 (20.27235°N, 102.75230°W); CH7 (20.32358°N, 102.78582°W); and CH8 (20.25016°N, 102.87219°W).

samples, 32 sediment samples, and a representative subset of fish specimens.

At each sampling site, surface water (~30 cm depth) and interstitial water (~30 cm above lakebed) were collected using a Van Dorn sampler. Prior to water collection, glassware was rinsed with high-purity methanol (>98%). For pesticide analysis, 2 L samples were transferred to clean glass bottles; AMPA (α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid) subsamples were preserved with EDTA (Ethylenediaminetetraacetic acid). Separate aliquots were prepared for water quality and heavy metals in appropriate containers. All water samples were kept at 4 °C and transported to the laboratory within 24 h. Benthic sediment was collected using a grab sampler (Ekman Dredge 77,933) retrieving the top ~5 cm; samples were homogenized and stored at 4 °C.

Four fish species were selected for evaluation in this study: the native Chapala catfish (*Ictalurus dugesii*) and three invasive species, the common carp (*Cyprinus carpio*), largemouth bass (*Micropterus salmoides*), and blue tilapia (*Oreochromis aureus*). Fish specimens were procured from small-scale commercial fishers on the morning of each campaign. Dorsal muscle tissue was excised from behind the head, and samples were pooled by species and size into composite samples (~500 g per composite), yielding approximately 11-13 pooled groups per campaign (Table S3). All samples were stored at 4 °C until analysis.

2.2. Physicochemical water quality analysis

At each site and during each sampling event, on-site measurements of key water quality parameters were conducted using a calibrated multi-parameter water quality probe (YSI EXO2, Xylem, United States). The probe was deployed at both surface and interstitial depths to measure the water temperature (°C); pH; dissolved oxygen (DO, mg/L); electrical conductivity (EC, μS/cm); and oxidation-reduction potential (ORP, mV). Additionally, water transparency was assessed using a Secchi disk at each site (Secchi depth in cm).

Water samples were analyzed for nitrates (NO_3^-), nitrites (NO_2^-), ammoniacal nitrogen (N–NH_4^+), total phosphorus (TP), sulfates (SO_4^{2-}), cyanides (CN^-), five-day biochemical oxygen demand (BOD_5), total Kjeldahl nitrogen (TKN), settleable solids (SS), total suspended solids (TSS), and chemical oxygen demand (COD) using NOM-001-SEMARNAT-2021 methods (SEMARNAT, 2022). For microbiological analysis, fecal coliforms (FC) were quantified using the Most Probable Number (MPN) method per Mexican Standard NMX-AA-042-SCFI-2015 (SE, 2016b). Results were reported as MPN/100 mL, with a method detection limit of $\geq$ 3.0 MPN/100 mL.

2.3. Pesticide analysis in water, sediment, and biota

All pesticide extractions and analyses were performed using internally validated methods according to Yüksel and Şen (2018) and Yüksel et al. (2023). A total of 17 pesticide compounds spanning various classes were targeted, including: four herbicides (atrazine, metolachlor, diuron, and glyphosate); one herbicide metabolite (aminomethylphosphonic acid (AMPA), the primary degradation product of glyphosate); one organophosphate (dimethoate); and one organochlorine derivative (dicofol); and a suite of ten insecticides/acaricides, comprising organochlorines (DDT and its metabolites DDE and DDD, ©-HCH (lindane), ®-HCH, ™-HCH, heptachlor, aldrin, endosulfan sulfate, and methoxychlor),. The selection of these compounds was based on their historical or current regional use, toxicity profiles, and persistence (Salazar-Flores et al., 2020; Silva-Madera et al., 2021).

Prior to extraction, water samples were filtered through 0.45-μm membranes, sediment samples were air-dried to a moisture content of $\leq$ 10%, and fish muscle samples (~500 g composite) were thawed, homogenized, and subsampled (15 g aliquot per composite).

For water, solid-phase extraction (SPE) was conducted on 200 mL of acidified sample (pH ~3) using an OASIS HLB cartridge (500 mg, 6 cc), washed, dried under nitrogen, and eluted with 1 mL of methanol containing 0.1% formic acid (v/v), then filtered through a 0.45 μm PTFE (polytetrafluoroethylene) membrane.

For sediment, liquid-liquid extraction (LLE) was performed on approximately 15 g of dried material using a 50:50 (v/v) MTBE:n-hexane mixture acidified with 0.1 N H_2SO_4, assisted by ultrasonication (15 min) and centrifugation (4000 rpm), after which the supernatant was evaporated to dryness under nitrogen and reconstituted in 1 mL methanol with 0.1% formic acid (v/v).

For fish tissue, dispersive solid-phase extraction (dSPE) was carried out using a commercial QuEChERS kit (Phenomenex roQ) with 15 g of homogenized tissue in acetonitrile containing 0.1% formic acid (v/v) and a salt packet (150 mg $MgSO_4$, 25 mg C18, 25 mg PSA), followed by centrifugation and 0.45 μm PTFE filtration.

Extracts were analyzed by UHPLC-MS/MS (Thermo Scientific TSQ Quantis) in multiple reaction monitoring mode. For each analyte, two transitions were monitored and reported for confirmation and quantification. The instrument was calibrated using a multi-point curve (minimum of 10 levels) with matrix-matched standards and quality control (QC) samples. Data were processed using Chromeleon™ (v7.3.2). Matrix spike recoveries ranged from 70 to 110%, meeting SANTE/2020/12,830 guidance (European Commission, 2021), with RSDs (Relative Standard Deviation) below 20%.

LODs (Limit of detection) and LOQs (Limit of quantification) were determined from signal-to-noise (S/N) ratios of 3:1 and 5:1, respectively: 0.10 ng/L for water and 0.10 ng/kg for sediment and fish, with higher LODs for AMPA (0.20 ng/L) (due to its higher polarity and ionization challenges). All reported concentrations were above the LOQ unless otherwise specified. Detailed QA/QC data are provided in Table S4.

2.4. Heavy metals analysis in water, sediment, and biota

The concentrations of heavy metals (As, Cd, Cu, Cr, Hg, Ni, Pb, Zn) in all matrices were determined using inductively coupled plasma-atomic emission spectrometry (ICP-AES; PerkinElmer Optima 8300DV, Shelton, CT, USA) in accordance with U.S. EPA Method 6010B (US EPA, 1996). These metals were selected based on their inclusion in Mexican regulations for water quality and environmental protection (NOM-001-SEMARNAT-2021 and CE-CCA-001/89) (SEMARNAT, 2022; SEDUE, 1989).

Sample digestion were performed following standard methods: EPA 3050B for sediments (U.S. EPA, 1996); NOM-117-SSA1-1994 for fish tissue (SSa, 1995); NMX-AA-051 for water (SE, 2016a). LODs/LOQs for water samples were: 0.010 mg/L for As, Cu, Cr, Ni, Pb, and Zn; 0.002 mg/L for Cd; and 0.001 mg/L for Hg. For sediment and fish tissue, LODs were: 1.0 mg/kg for As, Cu, Cr, Ni, Pb, and Zn; 0.25 mg/kg for Cd; and 0.1 mg/kg for Hg.

Quality control measures included the use of certified reference materials (NIST), with mean recoveries ranging from 85 to 115%. Reagent blanks were analyzed with each batch and were consistently below the respective LOQs for all metals. Analytical uncertainty, expressed as RSDs consistently below 20% for all metals and matrices.

2.5. Bioaccumulation factors

Bioaccumulation factors (WBF for water, SBF for sediment) were calculated following Pérez et al. (2021), based on average environmental concentrations in water and sediment above the LOD. Due to the high mobility of fish, environmental data were pooled across all stations and depths to generate monthly system-wide averages for water and sediment. Fish tissue concentrations were averaged per species per month.

2.6. Risk assessment

2.6.1. Environmental risk assessment

The chronic ecological risk to aquatic organisms was assessed by calculating ecological risk quotients (RQs) for pesticides detected in surface water and sediment, following the approach of Pérez et al. (2021). Briefly, RQ was derived as the ratio of measured environmental concentration (MEC) to predicted no-effect concentration (PNEC), where PNEC values were calculated from chronic NOEC (No Observed Effect Concentration) values using a standard assessment factor (AF) of 100 (OECD, 1982).

Chronic toxicity data (NOEC value) for algae (*Raphidocelis subcapitata*, 3-5 days), crustaceans (*Daphnia magna*, 21 days), and fish (*Danio rerio* and *Pimephales promelas,* 28 days) were sourced from the USEPA ECOTOX Knowledgebase (U.S. EPA, 2020). The sum of RQs (ΣRQ) was calculated to assess mixture risk. This summation approach, based on the concentration addition model, is widely used as a conservative first-tier tool in ecological risk assessments to identify potential risk hotspots and priority compounds requiring further investigation (U.S. EPA, 1998).

2.6.2. Human health risk assessment from fish consumption

Non-carcinogenic health risk from dietary pesticide exposure via fish consumption was assessed using the hazard quotient (HQ) method for adults and children.

Estimated daily intake (*EDI*, μg/kg•day) of each pesticide was calculated using Equation (1):

$$EDI = \frac{C_f \times FCR}{BW} \quad (1)$$

where C_f is the mean pesticide concentration in fish muscle (μg/kg), *FCR* is the daily fish consumption rate (g/day), and *BW* is the average body weight (kg).

The average fish consumption rate was set at 265 g/week for adults, based on national per capita consumption data (Estévez-Moreno and Miranda-de la Lama, 2022). For children, the consumption rate was estimated as 132.5 g/week, applying a 0.5 adjustment factor to the adult value in accordance with dietary patterns reported in the Mexican National Health and Nutrition Survey (Instituto Nacional de Salud Pública, 2019). Standard reference body weights of 70 kg (adults) and 20 kg (children) were applied, following the risk assessment protocol used by Arisekar et al. (2021).

For compounds not detected in fish tissue (concentrations below the LOD), EDI and HQ were not calculated. Instead, these compounds were assigned a concentration of zero for risk assessment purposes, following the conservative approach recommended by the U.S. EPA (1998) for screening-level assessments. This approach assumes that non-detects represent a negligible contribution to dietary exposure, which may underestimate risk but is consistent with standard practice in screening-level risk characterizations when detection frequencies are low (Arisekar et al., 2021; U.S. EPA, 1998).

Subsequently, the HQ was calculated for each pesticide as the ratio between the estimated daily intake (*EDI*) and the toxicological reference value, using Equation (2):

$$HQ = \frac{EDI}{ADI\ (or\ RfD)} \quad (2)$$

where *ADI* is the acceptable daily intake and *RfD* is the reference dose (μg/kg•day). Further details on the calculation procedure are provided in Supplementary Information (Table S10).

2.7. Statistical analysis

All statistical analyses were performed in R (version 4.5.1) using RStudio 2025.09.0. Data manipulation and visualization was conducted using the Tidyverse ecosystem (Wickham et al., 2019). Normality of data distribution was assessed using the Shapiro-Wilk test, and homoscedasticity was evaluated using Levene's test. Given the nature of the data and the small sample sizes ($n < 30$ per group), nonparametric methods were selected as the most appropriate analytical approach.

Outliers were identified using Walsh's test (EPA QA/G-9) and retained when environmentally justified (U.S. EPA, 2000). Seasonal and spatial differences in contaminant concentrations were each tested using the Kruskal-Wallis rank-sum test ($p < 0.05$) and Dunn's test with Benjamini-Hochberg corrections applied post-hoc. was applied when significant differences were detected (dunn.test package, Dinno, 2024), followed by letter grouping via multcompView (Graves et al., 2024) to indicate homogeneous subsets. All pairwise Dunn's test p-values were adjusted for multiple comparisons using the Benjamini-Hochberg false discovery rate correction. Seasonal and spatial factors were treated as independent grouping variables and tested separately. Only contaminants above their respective LOD were included in these comparisons, and boxplots were generated exclusively for those showing statistically significant differences, using ggplot2 (Wickham, 2016) with colorblind-friendly viridis palettes (Garnier et al., 2024).

Bioaccumulation factors values were $\log_{10}$-transformed after adding a small constant (0.001) to handle zero values, then used to generate heatmaps with hierarchical clustering (Euclidean distance, Ward's linkage) to group contaminants with similar bioaccumulation profiles across species, seasons, and sampling periods.

To test whether feeding guilds influence bioaccumulation according to contaminant physicochemical properties, WBF (Water bioaccumulation factor) and SBF (Sediment bioaccumulation factor) were compared between benthivorous (carp, catfish) and pelagic (tilapia, bass) species using Wilcoxon rank-sum tests for each contaminant separately. To account for multiple comparisons, p-values were adjusted using the Benjamini–Hochberg false discovery rate (FDR) correction.

To assess whether overall bioaccumulation profiles (considering all contaminants simultaneously) differed between feeding guilds, PERMANOVA (Permutational multivariate analysis of variance) was performed with 9999 permutations on Euclidean distance matrices using the vegan package (Oksanen et al., 2026). ANOSIM (Analysis of similarities) was additionally conducted as a complementary test to confirm the robustness of the PERMANOVA results. All multivariate analyses were performed separately for WBF and SBF.

To further explore the underlying patterns of contaminant bioaccumulation and identify the main drivers of variation among species and seasons, a principal component analysis (PCA) was performed including the seven detected pesticides and fish weight as an active variable using the factoextra package (Kassambara and Mundt, 2026) for visualization.

3. Results

3.1. Water quality characteristics

Seasonal and spatial trends in water quality were variable across the four sampling campaigns (Table 1). Several parameters exceeded regulatory thresholds (NOM-001-SEMARNAT-2021, CE-CCA-001/89, or ICA criteria) (SEMARNAT, 2022; SEDUE, 1989), particularly during the rain onset and cold-dry seasons. Interstitial dissolved oxygen indicated hypoxia near sediments across most seasons, while electrical conductivity, COD, TP, TSS, and TAN exceeded permissible limits in at least one season (Table 1). Fecal coliforms exceeded limits at multiple sites, with the highest values recorded during the rainy seasons (Table S5). All other parameters remained within regulatory limits.

Spatially, sites CH5, CH6, and CH7, located along the northern shore near the Lerma River inflow, consistently showed the highest contaminant concentrations (Table S5). In contrast, offshore sites CH2 and CH3 remained within regulatory limits for all parameters measured. Trophic indices classified the lake as hypertrophic based on TP and Secchi depth,

Table 1
Summary of mean seasonal water quality parameters for Lake Chapala.

Parameter	Unit	Rainy season onset	Rainy season cessation	cold-dry season	warm-dry season
DO Superficial	**%**	77.49	85.83	70.46	81.38
DO Interstitial	**%**	**24.59**	51.48	38.82	**18.06**
EC	**µS/cm**	**1079.28**	796.79	886.52	**1045.79**
pH	**-**	8.39	8.36	8.55	8.55
WT	**°C**	24.26	21.63	20.38	23.75
ORP	**mV**	−22.32	0.72	15.56	−65.28
Turbidity (Secchi)	**cm**	23.17	17.87	30.29	23.79
CN	**mg/L**	<0.025	<0.025	<0.025	<0.025
BOD	**mg/L**	2.42	2.41	2.39	2.09
COD	**mg/L**	**72.33**	27.38	**61.72**	34.6
TP	**mg/L**	**0.95**	**2.01**	**0.54**	**0.68**
TKN	**mg/L**	5.16	2.95	2.41	5.06
TAN	**mg/L**	<2.00	<2.00	**2.06**	<2.00
NO3	**mg/L**	1.26	1.00	1.39	0.17
NO2	**mg/L**	0.03	0.03	0.03	<0.01
Sulfate	**mg/L**	151.38	81.38	132.19	137.82
SS	**mg/L**	1.73	1.27	0.90	0.19
TSS	**mg/L**	**163.19**	**142.75**	**128.5**	65.19
FC	**MPN/100 mL**	795.44	578.06	168.19	89.44

*Values in bold indicate non-compliance with at least one of the following Mexican regulations: NOM-001-SEMARNAT-2021, CE-CCA-001/89, or the Water Quality Index (ICA) criteria. DO: dissolved oxygen; EC: electrical conductivity; WT: water temperature; ORP: oxidation-reduction potential; BOD: biochemical oxygen demand; COD: chemical oxygen demand; TP: total phosphorus; TKN: total Kjeldahl nitrogen; TSS: total suspended solids; FC: fecal coliforms.

although Chl-a indicated mesotrophic status (Table S6).

To evaluate the eutrophication process, the lake's condition was characterized using two common trophic indices: the Organisation for Economic Co-operation and Development (OECD) classification system and Carlson's Trophic State Index (TSI) (Table S6) (Carlson, 1977; OECD, 1982). Trophic indices classified the lake as hypertrophic based on TP (TSI TP: 94.9-113.8) and Secchi depth (TSI SD: 77.4-84.7), although chlorophyll-a (Chl-a) indicated mesotrophic status (TSI Chl: 44.4).

3.2. Pesticide and heavy metals occurrence in water and sediment

Across both the sediment and water matrices, the concentrations of lindane/©-HCH, ®-HCH, ™-HCH, heptachlor, dicofol, and aldrin were consistently below the method's LOD in all samples (<0.10 ng/L (kg)). Methoxychlor was detected in water but was present below quantifiable

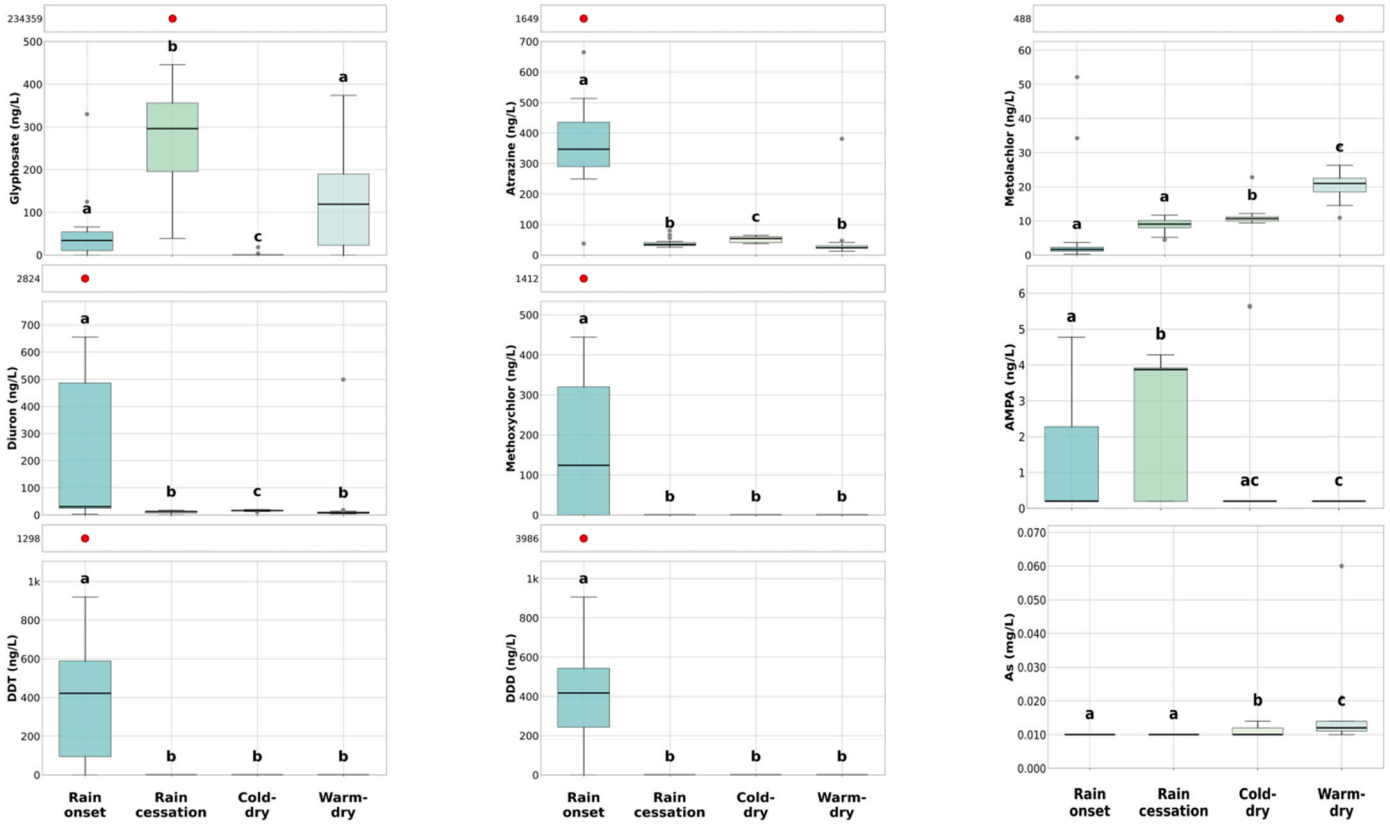


Fig. 2. Concentrations of detected water contaminants across the four sampling seasons with different letters indicating significant seasonal variation. Red points indicate outliers and boxplots show median and interquartile ranges. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

levels in sediment.

3.2.1. Water

In the water column, a pronounced seasonal trend was observed, with the highest detection frequency and maximum concentrations occurring during the rain-onset period. During this period, a complex mixture of legacy and current-use pesticides was detected across nearly all sites, including atrazine (up to 1649 ng/L), diuron (up to 2824 ng/L), methoxychlor (up to 1412 ng/L), DDT (up to 1298 ng/L), and DDD (up to 3986 ng/L) (Fig. 2).

Statistical analysis revealed significant seasonal differences for all evaluated compounds (Kruskal-Wallis, $p < 0.001$ for each; Fig. 2). During the rain-onset period, atrazine ($\chi^2 = 40.72$, df = 3, $p < 0.001$; Fig. 2), diuron ($\chi^2 = 36.75$, df = 3, $p < 0.001$; Fig. 2), and methoxychlor ($\chi^2 = 34.67$, df = 3, $p < 0.001$; Fig. 2) were all significantly higher than in other seasons. DDT and its metabolite DDD were detected at all of the sample sites only during the rain-onset period.

In contrast, glyphosate ($\chi^2 = 37.08$, df = 3, $p < 0.001$; Fig. 2) and AMPA ($\chi^2 = 17.87$, df = 3, $p < 0.001$; Fig. 2) peaked during the rain-cessation period. Metolachlor ($\chi^2 = 39.13$, df = 3, $p < 0.001$; Fig. 2) was significantly elevated during the warm-dry season. Endosulfan sulfate, however, showed no significant seasonal differences ($p > 0.05$), remaining relatively stable across all seasons (Table S7).

Arsenic (As) was significantly higher during the rain-onset period compared with all other seasons ($\chi^2 = 33.32$, df = 3, $p < 0.001$; Fig. 2). In contrast, zinc showed no significant seasonal differences (Table S7). Cd, Cu, Cr, Hg, Ni, and Pb fell below the LOD (<0.002-0.010 mg/L; Fig. 2). A notable exception, however, was observed at interstitial site 6 (CH6) during the warm-dry season, where analyses revealed measurable concentrations of As (0.012 mg/L), Cd (0.013 mg/L), Cu (0.073 mg/L), Cr (0.044 mg/L), Ni (0.037 mg/L), Pb (0.025 mg/L), and Zn (0.051 mg/L).

3.2.2. Sediment

The sediment compartment exhibited marked seasonal differences in contaminant concentrations (Kruskal-Wallis, $p < 0.05$ for most compounds; Fig. 3A). Three distinct seasonal patterns emerged.

Rain onset was characterized by the exclusive detection of the legacy pesticide DDT (5 out of 8 sites; 30.72–183.70 ng/kg), which was significantly higher than in all other seasons (Dunn test, $p < 0.01$; Fig. 3A). The metabolites DDD and DDE remained below the LOD across all sampling sites and seasons. This season also showed elevated concentrations of Ni (up to 12.28 mg/kg), which was significantly higher than during the cold-dry season ($p < 0.001$; Fig. 3A).

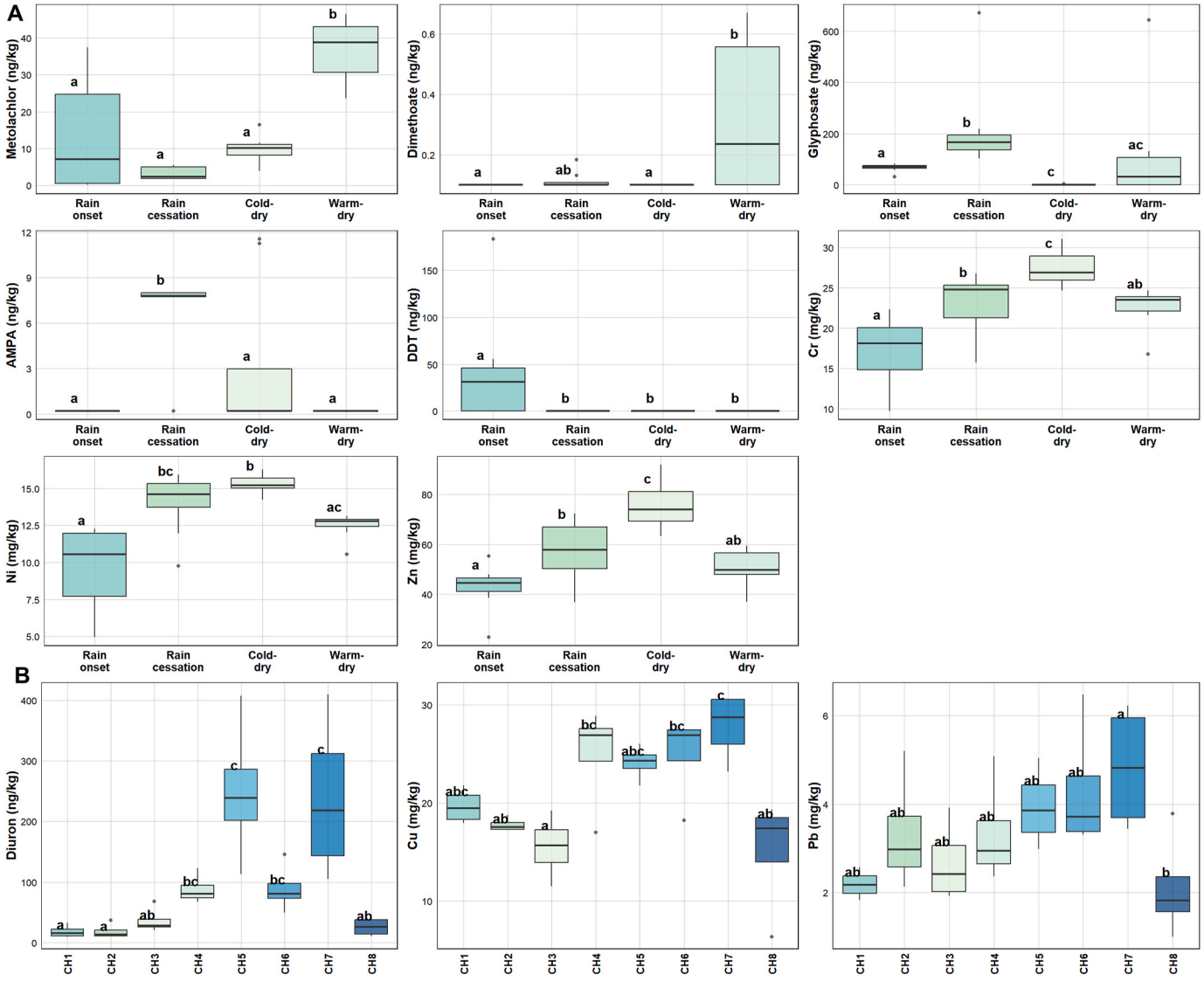


Fig. 3. Concentrations of sediment contaminants with significant seasonal (A) and spatial variation (B). Different letters indicate significant differences and boxplots show median and interquartile ranges.

Rain cessation was distinguished by for the highest levels of glyphosate (up to 671.23 ng/kg at CH6) and its metabolite AMPA (up to 8.02 ng/kg), both significantly elevated compared with the rain onset and cold-dry seasons ($p < 0.05$; Fig. 3A).

The cold-dry and warm-dry seasons showed contrasting metal dynamics. Cr peaked during the cold-dry season (up to 31.07 mg/kg), significantly exceeding all other seasons ($p < 0.05$). In contrast, Ni and Zn reached their highest concentrations during the warm-dry season (Ni: up to 13.16 mg/kg; Zn: up to 91.79 mg/kg), with Zn showing significant differences compared to rain onset and cold-dry ($p < 0.01$; Fig. 3A) seasons. Metolachlor and dimethoate also peaked during the warm-dry season (up to 46.48 ng/kg and 0.67 ng/kg, respectively), significantly exceeding rain onset and cold-dry seasons ($p < 0.05$; Fig. 3A).

Endosulfan sulfate and atrazine showed no significant seasonal variation, remaining detectable across all sites and seasons at low concentrations (Table S8). As, Cd, and Hg remained near or below the LOD across all samples.

Three contaminants exhibited significant spatial variability across the eight sampling sites (Kruskal-Wallis, $p < 0.05$). Diuron showed the strongest spatial differentiation ($\chi^2 = 25.90$, df = 7, $p < 0.001$; Fig. 3B), with elevated concentrations at CH4, CH5, and CH7 (Dunn test, $p < 0.05$; Fig. 3B). Cu also showed significant spatial variation ($\chi^2 = 19.70$, df = 7, $p = 0.006$; Fig. 3B), with the highest concentrations at CH4, CH5, CH6, and CH7. CH7 was notable for significantly higher Cu levels compared with CH2, CH3, and CH8 ($p < 0.01$). Pb exhibited significant spatial variability ($\chi^2 = 16.13$, df = 7, $p = 0.024$; Fig. 3B), with elevated levels at CH5, CH6, CH7, and CH8, peaking at CH7 (6.23 mg/kg). Pb at CH7 was significantly higher than at CH1 and CH3 ($p < 0.05$; Fig. 3B).

3.3. Pesticide and heavy metals bioaccumulation in fish

3.3.1. Pesticides

Fish muscle tissue displayed distinct contamination profiles across seasons (Fig. 4). All organochlorine pesticides (including methoxychlor, lindane/γ-HCH, β-HCH, δ-HCH, heptachlor, aldrin, dicofol, DDT, DDD, DDE) were below the LOD across all samples (<0.10 ng/kg).

Significant seasonal differences were observed for multiple residues. Atrazine (Kruskal-Wallis: $\chi^2 = 30.47$, df = 3, $p < 0.001$; Fig. 4) was significantly elevated during rain onset, with concentrations up to 112.30 ng/kg. Metolachlor ($\chi^2 = 28.56$, df = 3, $p < 0.001$; Fig. 4) showed significantly higher concentrations during the cold-dry season (up to 211.97 ng/kg). Diuron ($\chi^2 = 15.67$, df = 3, $p = 0.001$; Fig. 4) was significantly elevated during rain cessation, with concentrations up to 70.12 ng/kg. Dimethoate ($\chi^2 = 10.59$, df = 3, $p = 0.014$; Fig. 4) showed significantly higher concentrations during rain onset. Endosulfan sulfate

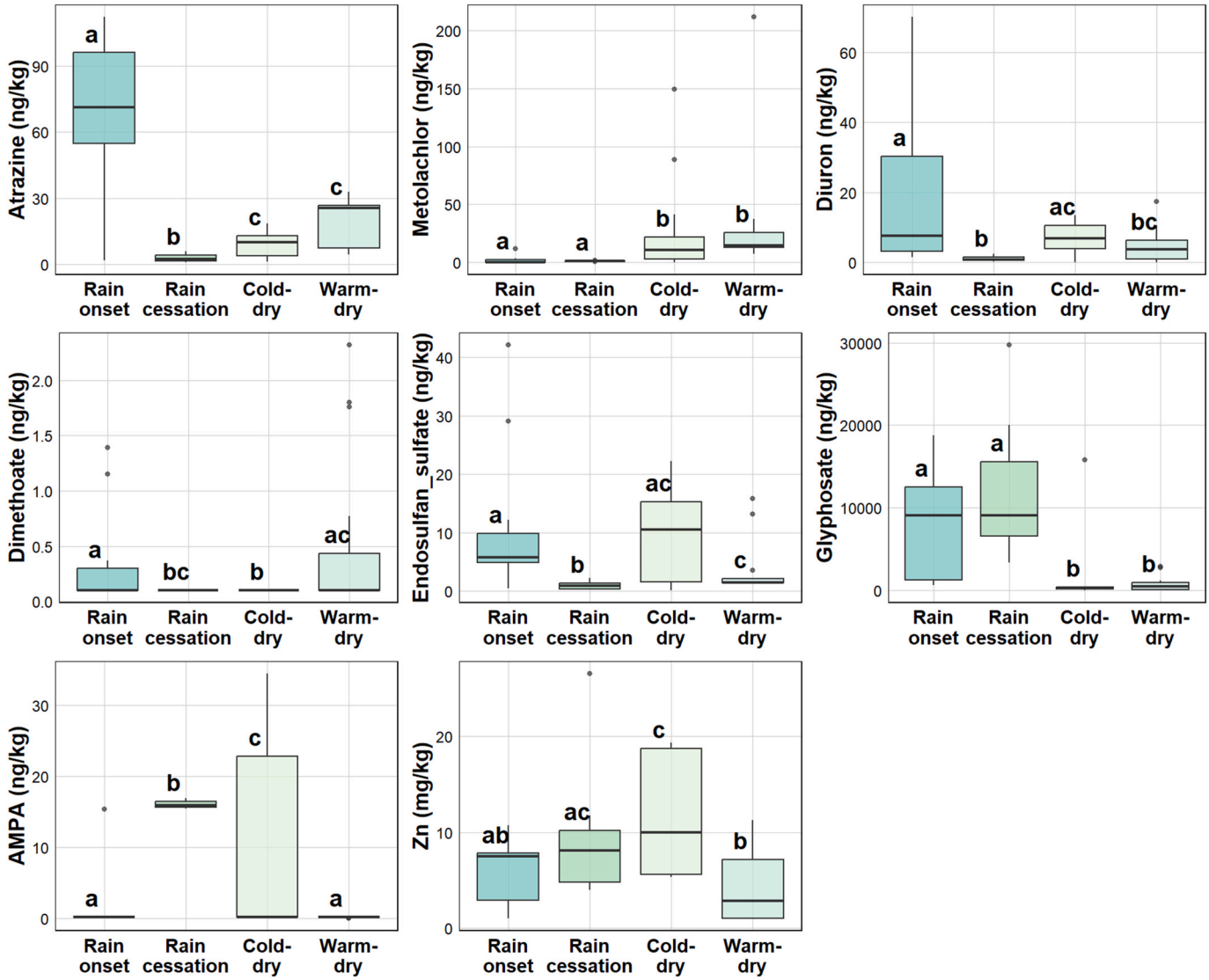


Fig. 4. Concentrations of contaminants found in fish. Different letters indicate significant differences and boxplots show median and interquartile ranges.

($\chi^2 = 21.46$, df = 3, $p < 0.001$; Fig. 4) was significantly elevated during rain cessation, with concentrations up to 42.13 ng/kg.

Glyphosate ($\chi^2 = 29.75$, df = 3, $p < 0.001$; Fig. 4) concentrations were significantly elevated during rain cessation (with concentrations up to 29,720.96 ng/kg) and rain onset (with concentrations up to 18,772.83 ng/kg). Dunn's test confirmed that both rain cessation and rain onset levels were significantly higher than the warm-dry season ($p < 0.001$ for both; Fig. 4) and cold-dry season ($p < 0.001$ for rain cessation; $p < 0.001$ for rain onset; Fig. 4).

AMPA ($\chi^2 = 27.82$, df = 3, $p < 0.001$; Fig. 4) was detected predominantly during rain cessation (with concentrations up to 34.48 ng/kg) and the cold-dry season (with concentrations up to 23.21 ng/kg), with significantly higher concentrations during the cold-dry season.

3.3.2. Heavy metals

Heavy metal concentrations in fish muscle were consistently low across all seasons. Zn was the only metal detected above trace levels, while As, Cd, Cu, Cr, Hg, Ni, and Pb remained near or below LOD in all samples (Table S9).

Zn showed significant seasonal differences (Kruskal-Wallis: $\chi^2 = 12.24$, df = 3, $p = 0.007$; Fig. 4). The highest zinc levels were recorded during the warm-dry season (with concentrations up to 7.78 mg/kg) and the rain cessation season (with concentrations up to 26.60 mg/kg).

3.3.3. Bioaccumulation patterns

Carp (*Cyprinus carpio*) exhibited the highest zinc levels (ranging from 5.27 to 26.6 mg/kg), followed by catfish (*Ictalurus dugesii*) (with concentrations of 1.00–11.67 mg/kg), then blue tilapia (*Oreochromis aureus*) (with concentrations 1.00-5.64 mg/kg), and largemouth bass (*Micropterus salmoides*) (with concentrations of 1.65-5.78 mg/kg) (Fig. 5). Atrazine and metolachlor consistently showed low concentrations across all species. Glyphosate and zinc consistently exhibited the highest bioaccumulation factors among all analytes across most species and seasons (WBF: 0.71-263.39 for glyphosate; 73.49-1529.09 for zinc; Fig. 5), indicating a strong accumulation potential for both compounds in fish tissue. This pattern was further supported by the bioaccumulation factor analysis, which revealed pronounced glyphosate accumulation from the water phase (WBF) rather than from sediment (SBF) across all species.

Multivariate analyses revealed that WBF differed significantly between benthivorous and pelagic species (PERMANOVA: $F = 2.3049$, $R^2 = 0.0458$, $p = 0.0021$; ANOSIM: $R = 0.1829$, $p = 0.0012$). No significant differences were detected for SBF (PERMANOVA: $F = 0.6842$, $R^2 = 0.0141$, $p = 0.6434$; ANOSIM: $R = -0.0009$, $p = 0.4225$). Univariate comparisons revealed that zinc was the only contaminant showing significantly higher bioaccumulation in benthivorous species ($W = 525$, $p < 0.001$). Numerical trends were observed for endosulfan (ratio = 1.95; $W = 359.5$, $p = 0.243$) and glyphosate (ratio = 8.22; $W = 289.5$, $p = 0.843$), both showing higher mean values in benthivores. However, the high within-group variability limited statistical power, preventing these differences from reaching significance.

PCA on the seven detected pesticides explained 56.6% of the total variance (PC1: 36.4%, PC2: 20.2%). PC1 was positively correlated with atrazine, diuron, and endosulfan sulfate; PC2 defined an independent

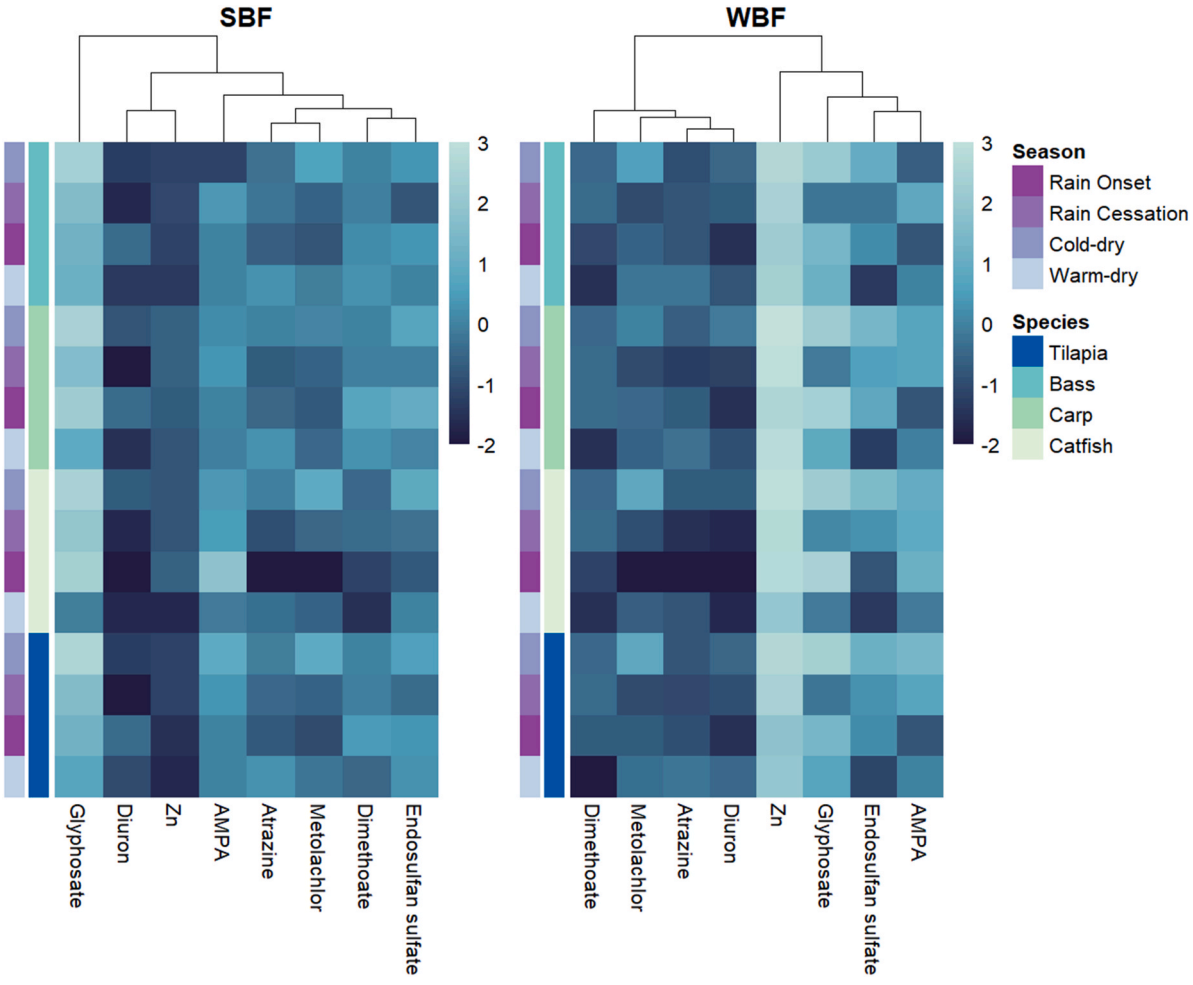


Fig. 5. Heatmap of $\log_{10}$-transformed bioaccumulation factors for contaminants in fish muscle tissue from Lake Chapala. SBF: Sediment-based bioaccumulation factor; WBF: Water-based bioaccumulation factor.

gradient between glyphosate (positive) and metolachlor (negative). Fish weight showed minimal contribution to PC1 (loading = 0.14) and PC2 (loading = 0.28), suggesting that body mass was not a primary determinant of the observed contamination patterns.

3.4. Ecological and human health risk assessment

Within the preliminary toxicological health assessment (consumption of contaminated product), minimal risks were identified for the quantified pesticide concentrations in fish; no significant threat was identified when comparing pesticide concentrations to non-carcinogenic endpoints (HQ < 1 for all species and seasons). The preliminary assessment relied on accurate fish consumption data and used the estimated daily intake and reference doses (Table S10).

Ecological risk (RQ) depended on season and chemical mass. When the human health risk assessment was expanded to consider the sum of contaminants present, the rain onset season had the highest ΣRQ value of 935.87, followed by warm-dry (10.24), cold-dry (4.38), and rain cessation (3.21) seasons. The ΣRQ was primarily driven by four contaminants: DDD (RQ = 693.9); diuron (RQ = 167.1); methoxychlor (RQ = 25.8); and atrazine (RQ = 4.69) (Table 2).

4. Discussion

Collectively, the seasonal water quality patterns in Lake Chapala were indicative of a system strongly influenced by hydrological cycles and anthropogenic pressures. Through intensive sampling and non-targeted screening our results demonstrate the benefit of a multi-matrix approach to capture the full chemical exposome with epidemiological relevance relating to CKD. We demonstrate how rainfall events combined with presence of both legacy and current-use agrochemicals and matrix interactions facilitating metal mobilization create a

Table 2
Risk Quotients (RQs) and sum of risk quotients (ΣRQ) for detected pesticides in Lake Chapala surface water across four seasonal sampling periods. Values represent seasonal means from eight sampling stations. RQs were calculated as the ratio of measured environmental concentration to predicted no-effect concentration derived from chronic toxicity data for algae, Daphnia, and fish. ΣRQ values ≥ 1 indicate high ecological risk.

Pesticide	RQ Rain Onset	RQ Rain Cessation	RQ Cold-dry	RQ Warm-dry
Atrazine	4.6879	0.3981	0.5145	0.4844
Metolachlor	0.02724	0.03476	0.0454	0.20092
Diuron	167.10227	2.50455	3.69773	8.99773
Methoxychlor	25.823	0.01	0.01	0.01
Dimethoate	0.00385	0.00065	0.00075	0.06005
Endosulfan sulfate	0.03315	0.00435	0.00598	0.37804
Glyphosate	0.00053	0.14894	0.00002	0.0013
AMPA	0.00013	0.00028	0.00009	0.00002
DDT	44.295	0.01	0.01	0.01
DDD	693.89	0.1	0.1	0.1
DDE	0.00899	0.0001	0.0001	0.0001
∑RQ	935.87206	3.21173	4.38457	10.24256

hazardous water-food nexus which threatens One Health of humans and nature. As accumulation patterns were not related to fish body mass, fish species feeding ecology may be useful to triage contamination risk rather than invasion status (South et al., 2025; Franco et al., 2025). From a regulatory perspective our data shows that Lake Chapala receives discharges that do not comply with federal standards, compromising its designated uses under Mexican law, which include fishing, recreation, and as a water supply source for downstream agricultural and urban areas. Consequently, the lake cannot be considered a "healthy" aquatic ecosystem under Mexican regulatory frameworks, particularly during the rain onset and cold-dry seasons when multiple stressors converge.

Overall water quality in Lake Chapala was classified as moderate to poor throughout the study period according to Mexican regulations (NOM-001-SEMARNAT-2021, CE-CCA-001/89, and the Water Quality Index, ICA) (SEMARNAT, 2022; SEDUE, 1989). The seasonal patterns and magnitude of contamination observed in this study were consistent with previous investigations in the Lerma-Chapala basin. Murillo-Delgado et al. (2021) had documented similar first-flush effects for nutrients and suspended solids, while Vallejo-Rodríguez et al. (2017) reported FC concentrations exceeding 10,000 MPN/100 mL at the Lerma River inflow, thus confirming this river as the primary chronic pollution source. The rain onset period consistently exhibited the poorest conditions. This trend is related to biogeochemical processes such as negative ORP, whereby reducing conditions have been identified to promote the reductive dissolution of metal oxides in sediments Ying et al., 2025; Ramírez-Sánchez and Fajardo-Montiel, 2024), and may trigger sulfate reduction, as suggested by seasonal sulfate peaks (up to 151.38 mg/L during rain onset), a process linked to phosphorus release (Ramírez-Sánchez and Fajardo-Montiel, 2024). Furthermore, first-flush mobilization of accumulated watershed contaminants was likely exacerbated by evaporation and low flow in the warm-dry season. (Vallejo-Rodríguez et al., 2017; Murillo-Delgado et al., 2021; Ramírez-Sánchez and Fajardo-Montiel, 2024). Lower COD during the warm-dry season indicated that higher temperatures enhanced microbial activity and photochemical oxidation (Ramírez-Sánchez and Fajardo-Montiel, 2024). The decoupling between COD and BOD, where elevated COD coincided with norm-compliant BOD, indicated a substantial proportion of refractory organic compounds, consistent with the Lerma-Chapala basin receiving an estimated 424,260 t/yr of COD compared to 130,500 t/yr of BOD from industrial and urban sources (de la Mora-Orozco et al., 2018). Persistent interstitial hypoxia, combined with elevated fecal indicators, highlighted a system under dual pressure from organic enrichment and microbial contamination (Vallejo-Rodríguez et al., 2017).

Trophic status of the lake directly influences environmental fate and bioavailability of co-occurring pollutants. In eutrophic and hypertrophic systems, high primary productivity, organic matter sedimentation and periodic hypoxia alter pollutant dynamics through sorption and reducing conditions promoting mobilization of metals from sediments (Neverova-Dziopak and Kowalewski, 2018) and transformation of pesticide metabolites (Le Cor et al., 2021). A contradiction was observed between trophic indices: while total phosphorus (TSI TP: 94.9-113.8) and Secchi depth (TSI SD: 77.4-84.7) indicated hypertrophic status, chlorophyll-a (TSI Chl: 44.4) indicated mesotrophic conditions. This pattern is a classic feature of shallow, turbid lakes like Lake Chapala, where severe light limitation from non-algal turbidity disrupted the linear relationship between high nutrient concentrations and algal biomass. This dynamic has been well-documented in this ecosystem, as confirmed by Membrillo-Abad et al. (2016), who documented a TSI value of 42 under OECD criteria, consistent with mesotrophic classification (OECD, 1982). Standard trophic indices were challenged in this light-limited system, where inorganic suspended sediments attenuated the light and constrained phytoplankton productivity despite high nutrient availability (Membrillo-Abad et al., 2016; Lara-González et al., 2024). Hypereutrophic conditions thus modify the persistence, distribution, and ecological risk of chemical contaminants in Lake Chapala. This was obvious in the near-exclusive partitioning of trace metals into lake sediments, consistent with lentic systems at high pH, explained why only Zn and As were consistently detectable in the water column, where particulate adsorption dominated over dissolution, reducing aqueous bioavailability (Tian et al., 2020; Alvarado et al., 2021).

The apparent absence of legacy organochlorines (including lindane, heptachlor, and aldrin) could be interpreted as a potentially positive finding; this result warranted cautious interpretation. Ecotoxicological benchmarks for chronic, sub-lethal effects of these compounds, particularly in environmental mixtures, often were lacking or exceeded concentrations that challenged even the most sensitive analytical methods. These LOD were several orders of magnitude above known sub-lethal effect thresholds for some sensitive organisms (Rico et al., 2025). This finding aligned with the national monitoring gap identified by de Anda et al. (2024), who reported that routine water quality assessments in Mexico (RENAMECA) consistently omitted organic contaminants, and with Garcia-Hernández et al. (2021), who documented the agricultural ban on DDT since 1991. However, our detection of DDT and its metabolites exclusively during rain onset partially diverged from Garcia-Hernández et al. (2021), who had documented more persistent sedimentary residues in other Mexican basins, suggesting that Lake Chapala's watershed functions primarily as a seasonal reservoir rather than a continuous source of legacy pesticides. Therefore, these results did not entirely preclude chronic ecological risks, highlighting the importance of continued development of even more sensitive analytical techniques and long-term, integrated ecotoxicological assessments.

Pesticides showed differing transport and fate patterns, which can be explained by the fundamental physicochemical properties of the detected pesticides. Legacy organochlorines (including DDT, methoxychlor, and HCH isomers) are governed primarily by hydrophobicity and the tendency to partition into organic phases such as sedimentary organic matter and biological lipids (Arp et al., 2017; Sigmund et al., 2022). These are more likely to bind to soils and sediments before mobilization during first-flush events. Glyphosate and its metabolite AMPA are pH-sensitive compounds which, under the lake's alkaline conditions (pH 8.36-8.55), these molecules existed predominantly as anions or zwitterions (Trinh a Schäfer, 2023). Strong sediment adsorption to soil minerals explains lower likelihood of transport from flushing (Silva Madera et al., 2021). Methoxychlor was only found in the water column, which illustrates redox-driven compartmentalization. While surface water exhibited suboxic to weakly oxidizing conditions, interstitial water was consistently more reducing, facilitating anaerobic degradation pathways such as reductive dechlorination (Boonman et al., 2024). Thus, we show that ORP values (−65.28 mV) during the warm-dry season were sufficiently reducing to promote methoxychlor degradation in benthic compartments in a tropical shallow lake. Detection of DDT/DDD only during the rain onset season suggests seasonal run-off from the watershed rather than legacy sedimentary deposits within the lake. This is supported by concurrent peaks in COD and TSS, consistent with first-flush mobilization of contaminated soils (Wang et al., 2025). The marked seasonal peak in pesticide concentrations during rain onset aligns with regional agricultural practices corresponding to the pre-planting season, when pesticides are documented to be intensively applied (Lozano-Kasten et al., 2021; Schönenberger et al., 2022; de Anda et al., 2022). This mirrors findings in Laguna del Cisne (Uruguay), where DDT exhibited the highest ecological risk among all pesticides evaluated (RQ up to 230), driven by agricultural runoff during peak application seasons (Rodríguez-Bolaña et al., 2024).

The sediment of the lake acts as an integrating sink which scavenged pollutants from the water column through sorption and settling, while also being subject to redox reactions and microbial transformations that influenced contaminant fate (Chiaia-Hernández et al., 2022). The localized detection of multiple metals in interstitial water at CH6 during the warm-dry season (with Cd and Pb exceeding NOM-001-SEMARNAT-2021 limits) demonstrated that this sedimentary sink was not permanent, but rather locally susceptible to remobilization

(SEMARNAT, 2022). This phenomenon was supported by Ramírez-Sánchez and Fajardo-Montiel (2024), who documented similar metal mobilization under negative ORP conditions in Lake Chapala. Our findings further identified CH6 as a specific hotspot where hydrological and redox conditions converged, a pattern previously undocumented in this lake. The site's proximity to the Santiago-Zula River confluence likely creates a hydrodynamic disturbance that resuspends and transports sediment-bound metals, confirming that hydrodynamic energy could override adsorptive retention (Ying et al., 2025).

There was a clear differential bioaccumulation of pesticides and trace metals among fish species, which was linked directly to their ecological niches influencing contaminant uptake, storage, and elimination (Nyeste et al., 2024; Alvarado et al., 2021). Stong et al. (2013) found high concentrations of mercury in carp especially which was not registered in our data. Our Zn concentrations in carp (5.27-26.6 mg/kg) and catfish (1.0–11.67 mg/kg) were generally lower than those reported by Alvarado et al. (2021) for the same benthic species in Lake Chapala (carp: 8.84-16.05 mg/kg; catfish: 5.66-12.64 mg/kg). Together these changes indicate reduced metal loading in recent years. This pattern of higher Zn accumulation in benthic species compared with pelagic feeders was consistent with the expected role of feeding ecology in governing contaminant uptake (Nyeste et al., 2024). Atrazine and metolachlor showed consistently low bioaccumulation potential, consistent with their relatively low log Kow values (~2.6 and ~3.05), limiting partitioning into lipid-rich tissues (Gianni et al., 2022; do Carmo et al., 2023). Benthic feeders are more likely to accumulate glyphosate, AMPA, and endosulfan sulfate due to grubbing behaviours resuspending sediment particles (Cornish et al., 2023; Bonansea et al., 2018). Glyphosate's highwater solubility and anionic character ought to predicate low bioaccumulation potential, however, co-occurrence with dissolved metals such as zinc explain this trend. The formation of stable, soluble complexes between glyphosate/AMPA and divalent metal cations (Zn^{2+}, Ca^{2+}) in the water column could neutralize their charge, increase effective lipophilicity, and thereby facilitate membrane uptake (Mertens et al., 2018; Mohy-Ud-Din et al., 2023). This inference is supported by a positive correlation between glyphosate and zinc concentrations in fish tissue, the pronounced dominance of WBF over SBF, and the preferential accumulation in benthic feeders. The mechanism behind glyphosate–metal co-exposure has been linked to renal tubular damage in agricultural communities (Jayasumana et al., 2015; Wanigasuriya et al., 2011). Future mechanistic work should focus on experimental validation through controlled exposure trials with metal-glyphosate complexes at environmentally relevant concentrations and speciation modelling.

The elevated contaminant burden in the native Chapala catfish was of conservation concern, as this species is in decline due to habitat alteration and competition with invasive species (Juárez-Aguilar and López-Gómez, 2024). Chronic exposure to the reported levels of Zn, glyphosate, and AMPA may represent an additional stressor, alongside climate change and invasive species, that can impair reproduction, immune function, or foraging efficiency (Ray and Shaju, 2023). The concurrent detection of elevated zinc and glyphosate/AMPA in these fish was consistent with an agricultural chemical signature, as zinc is a common additive in livestock feed and phosphate fertilizers that contributed to adjacent water bodies through runoff (Jayasumana et al., 2015; Mertens et al., 2018; Bonansea et al., 2018).

The future of Lake Chapala and the health of the communities which rely on it depends on developing proactive regulation of contaminants entering the watershed, integrative policy recommendations regarding fish consumption and aquaculture in the lake, and the capacity of the regional government to ensure clean water access. These data established a critical baseline for the lake and provided actionable evidence to regulate glyphosate, diuron, and legacy organochlorines. Mexico's monitoring framework needs to expand to include organic micropollutants and interstitial water at river confluences, and a glyphosate phase-out should be accelerated with post-ban monitoring. Communities should be encouraged to promote consumption of pelagic fish. However, the most predominantly available are non-native species which contribute to the massive population declines of the endemic *charales* (Chirostoma spp.). These are small sardines which have low bioaccumulation and high nutrient content, thus would provide an alternative protein source if the fishery was to suitably recover (Moncayo-Estrada and Buelna-Osben, 2001; Heilpern et al., 2025). While the onus is on government authorities to provide clean water, this should be combined with small scale interventions co-created with communities as alternatives.

4.1. Hydrological drivers of water quality and contaminant dynamics

Overall water quality in Lake Chapala was classified as moderate to poor throughout the study period according to Mexican regulations (NOM-001-SEMARNAT-2021, CE-CCA-001/89, and the Water Quality Index, ICA) (SEMARNAT, 2022; SEDUE, 1989). The sustained exceedances documented for multiple parameters, including interstitial DO, TP, FC, TSS, COD, and TAN, indicate non-compliance with at least one of the aforementioned regulations. The rain onset period consistently exhibited the poorest conditions. From a regulatory perspective, these exceedances indicate that Lake Chapala receives discharges that do not comply with federal standards, compromising its designated uses under Mexican law, which include fishing, recreation, and as a water supply source for downstream agricultural and urban areas. Consequently, the lake cannot be considered a “healthy” aquatic ecosystem under Mexican regulatory frameworks, particularly during the rain onset and cold-dry seasons when multiple stressors converge. These findings are consistent with previous assessments of Lake Chapala's water quality, which have similarly reported eutrophic conditions with elevated nutrient concentrations and reduced transparency (Membrillo-Abad et al., 2016; Lara-González et al., 2024).

The negative ORP values during the rain onset (−22.32 mV) and warm-dry season (−65.28 mV) were the primary biogeochemical drivers explaining metal mobilization at site CH6 and the differential persistence of organochlorines like methoxychlor. Reducing conditions have been identified to promote the reductive dissolution of metal oxides in sediments (Ying et al., 2025; Ramírez-Sánchez and Fajardo-Montiel, 2024), and may trigger sulfate reduction, as suggested by seasonal sulfate peaks (up to 151.38 mg/L during rain onset), a process linked to phosphorus release (Ramírez-Sánchez and Fajardo-Montiel, 2024).

EC exceedances during both rain onset and warm-dry seasons arose from fundamentally different mechanisms. The early rainy season peak reflects reflected first-flush mobilization of accumulated salts from the watershed (Murillo-Delgado et al., 2021), while the warm-dry season exceedance results resulted from evaporative concentration under reduced inflow conditions (Ramírez-Sánchez and Fajardo-Montiel, 2024). Later rainfall diluted ionic concentrations, returning conductivity to acceptable levels, demonstrating that exceedances could signal either watershed flushing or hydrological stress, with different management implications.

Elevated COD at rain onset was attributable to first runoff events transporting accumulated oxidizable substances from the watershed, consistent with previous studies identifying the Lerma River basin as a significant contributor to water quality fluctuations (Murillo-Delgado et al., 2021). The persistence of elevated COD during the cold-dry season suggested internal lake processes, while lower COD during the warm-dry season indicated that higher temperatures enhanced microbial activity and photochemical oxidation (Ramírez-Sánchez and Fajardo-Montiel, 2024). The decoupling between COD and BOD, where elevated COD coincided with norm-compliant BOD, indicated a substantial proportion of refractory organic compounds, consistent with the Lerma-Chapala basin receiving an estimated 424,260 t/yr of COD compared to 130,500 t/yr of BOD from industrial and urban sources (de la Mora-Orozco et al., 2018).

The persistent hypoxia in interstitial waters, despite normoxic

surface conditions, demonstrated that surface monitoring alone was inadequate to assess oxygen availability throughout the lake system. This vertical gradient poses profound implications for benthic habitat quality and nutrient cycling, as prolonged low-oxygen conditions near sediments could alter biogeochemical cycles and release stored nutrients back into the water column (Murillo-Delgado et al., 2021).

The extreme FC contamination during rain onset, with multiple sites reaching 2400 MPN/100 mL, evidenced a first-flush phenomenon where initial rainfall mobilized accumulated fecal matter from the catchment, creating a compounded microbial risk superimposed on chronic high-level pollution from the Lerma River (Vallejo-Rodríguez et al., 2017). This event was directly relevant to public health, as diarrhea mortality rates in lakeside communities were three times the national average (Smith et al., 2020). Beyond immediate risks, chronic exposure to sewage-impacted freshwater of the kind documented here had been associated with renal histopathological damage in resident fish, including tubular necrosis, interstitial congestion, and tubular degeneration (Hussain et al., 2019; Camargo and Martinez, 2007). Whether analogous exposure pathways affect human health in lakeside communities remains to be determined. Nevertheless, epidemiological studies in regions with high rates of chronic kidney disease of unknown origin have documented pervasive fecal contamination in groundwater, with over 90% of wells showing total coliforms and half testing positive for *Escherichia coli* in Sri Lanka's CKDu endemic zone (Imbulana et al., 2020). Although these studies do not establish a direct statistical correlation with CKDu prevalence, they confirm chronic exposure to microbiologically unsafe water, frequently alongside other potential renal stressors such as elevated hardness and magnesium (Imbulana et al., 2020). The multistressor environment we documented in Lake Chapala, combining microbial contamination, elevated hardness, and trace metal co-occurrence, bears similarities to these settings. We offer this convergence as hypothesis generating rather than diagnostic, and we emphasize that establishing direct links between environmental contamination and human health outcomes would require dedicated epidemiological and toxicological studies beyond the scope of this work.

The seasonal patterns and magnitude of contamination observed in this study were consistent with previous investigations in the Lerma-Chapala basin. Murillo-Delgado et al. (2021) had documented similar first-flush effects for nutrients and suspended solids, while Vallejo-Rodríguez et al. (2017) reported FC concentrations exceeding 10,000 MPN/100 mL at the Lerma River inflow, thus confirming this river as the primary chronic pollution source.

Collectively, the seasonal water quality patterns in Lake Chapala were indicative of a system strongly influenced by hydrological cycles and anthropogenic pressures. During rainfall periods, there were conductivity and nutrient peaks, which illustrated watershed flushing, whereas in the warm-dry season, concentrations resulted from evaporation and internal recycling. The dissociation between COD and BOD indicated a predominance of refractory organic matter, likely derived from industrial and agricultural sources. Persistent interstitial hypoxia, combined with elevated fecal indicators, highlighted a system under dual pressure from organic enrichment and microbial contamination, posing implications for both ecosystem health and human use.

4.2. Trophic status and its implications for pollutant dynamics

Determining the lake's trophic status was ecologically critical, as it directly influenced the environmental fate and bioavailability of co-occurring pollutants. In eutrophic and hypertrophic systems, high primary productivity, organic matter sedimentation, and periodic hypoxia can significantly have altered pollutant dynamics: there was enhanced sorption of hydrophobic pesticides and metals to suspended particles may occur, while reducing conditions in sediments promoted mobilization of certain metals (Neverova-Dziopak and Kowalewski, 2018) and transformation of pesticide metabolites (Le Cor et al., 2021).

A contradiction was observed between trophic indices: while total phosphorus (TSI TP: 94.9-113.8) and Secchi depth (TSI SD: 77.4-84.7) indicated hypertrophic status, chlorophyll-a (TSI Chl: 44.4) indicated mesotrophic conditions. This pattern is a classic feature of shallow, turbid lakes like Lake Chapala, where severe light limitation from non-algal turbidity disrupted the linear relationship between high nutrient concentrations and algal biomass. This dynamic has been well-documented in this ecosystem, as confirmed by Membrillo-Abad et al. (2016), who documented a TSI value of 42 under OECD criteria, which is consistent with mesotrophic classification (OECD, 1982). Standard trophic indices were challenged in this light-limited system, where inorganic suspended sediments attenuated the light and constrained phytoplankton productivity despite high nutrient availability (Membrillo-Abad et al., 2016; Lara-González et al., 2024).

The persistent hypertrophic conditions in Lake Chapala therefore reflected not only nutrient over-enrichment but also a biogeochemical environment that modifies the persistence, distribution, and ecological risk of chemical contaminants.

4.3. Pesticide and heavy metals occurrence, transport, and fate in water and sediments

4.3.1. Pesticides

The apparent absence of legacy organochlorines (e.g., including lindane, heptachlor, and aldrin) could be interpreted as a potentially positive finding; this result warranted cautious interpretation. It was critical to acknowledge that ecotoxicological benchmarks for chronic, sub-lethal effects of these compounds, particularly in complex environmental mixtures, are often lacking or exceeded at concentrations that challenged even the most sensitive analytical methods. These LOD were several orders of magnitude above known sub-lethal effect thresholds for some sensitive organisms (Rico et al., 2025). This finding aligned with the national monitoring gap identified by de Anda et al. (2024), who reported that routine water quality assessments in Mexico, conducted by the National Water Quality Monitoring Network (RENAMECA), consistently omitted organic contaminants, and with García-Hernández et al. (2021), who documented the agricultural ban on DDT since 1991. However, our detection of DDT and its metabolites exclusively during rain onset partially diverged from García-Hernández et al. (2021), who had documented more persistent sedimentary residues in other Mexican basins, suggesting that Lake Chapala's watershed functions primarily as a seasonal reservoir rather than a continuous source of legacy pesticides. Therefore, these results did not entirely preclude chronic ecological risks, highlighting the importance of continued development of even more sensitive analytical techniques and long-term, integrated ecotoxicological assessments.

The selective presence of methoxychlor in the water column, contrasted with its absence in sediment and biota, illustrated redox-driven compartmentalization. While surface water exhibited suboxic to weakly oxidizing conditions, interstitial water was consistently more reducing, facilitating anaerobic degradation pathways such as reductive dechlorination (Boonman et al., 2024). As a result, methoxychlor entering porewater was degraded before accumulating in sediments or benthic organisms, while it persisted in the oxygenated water column where oxidizing conditions inhibited reductive transformations. This redox-driven partitioning was consistent with the findings of Boonman et al. (2024) in anaerobic sediment systems, and our results extended this mechanism to a tropical, shallow lake context, indicating that ORP values (−65.28 mV) during the warm-dry season were sufficiently reducing to promote methoxychlor degradation in benthic compartments.

The exclusive detection of DDT and its metabolites during rain onset argued against steady release from legacy sedimentary deposits within the lake, indicating instead a seasonal runoff-driven input from the watershed. This interpretation was reinforced by concurrent peaks in COD and TSS, consistent with first-flush mobilization of contaminated soils (Wang et al., 2025). The marked seasonal peak in pesticide

concentrations during rain onset aligned with regional agricultural practices corresponding to the pre-planting season, when pesticides were intensively applied (Lozano-Kasten et al., 2021), and it strongly supported a runoff-driven input mechanism where first rainfall events mobilized recently applied agrochemicals (Schönenberger et al., 2022; de Anda et al., 2022). This seasonal pattern agreed with Schönenberger et al. (2022), who had documented similar first-flush pesticide mobilization in agricultural watersheds, and with Lozano-Kasten et al. (2021), who had identified pre-planting applications as the primary driver of seasonal contamination peaks in the Lerma-Chapala basin.

These contrasting transport patterns could be explained by the fundamental physicochemical properties of the detected pesticides, which fell into two distinct categories. Legacy organochlorines (including DDT, methoxychlor, and HCH isomers) were neutral organic compounds whose environmental behavior was not pH-dependent within the typical aqueous range (Arp et al., 2017); their distribution was governed primarily by hydrophobicity and the tendency to partition into organic phases such as sedimentary organic matter and biological lipids (Sigmund et al., 2022). This pattern explained their affinity for sediments and their persistence in the watershed, where they remained bound to soils until mobilized by erosion during first-flush events.

In contrast, glyphosate and its metabolite AMPA were pH-sensitive compounds. Under the lake's alkaline conditions (pH 8.36-8.55), these molecules existed predominantly as anions or zwitterions (Trinh and Schäfer, 2023). Their anionic character, combined with strong adsorption to soil minerals rather than organic matter, explained their delayed transport via erosion processes rather than immediate mobilization with first-flush runoff. This delayed transport mechanism differed from the behavior of more soluble herbicides like atrazine and diuron, which mobilized readily with initial surface runoff, and in agreement with Silva Madera et al. (2021), who reported that up to 88.1% of applied glyphosate could be retained in the surface soil layer, making its transport more closely associated with erosion processes than with first-flush events. This distinction between contaminant classes was essential for understanding their differential transport, fate, and eventual bioavailability in the Lake Chapala system.

4.3.2. Heavy metals

The near-exclusive partitioning of trace metals into lake sediments, consistent with lentic systems at high pH, explained why only Zn and As were consistently detectable in the water column, where particulate adsorption onto suspended particles dominated over dissolution, reducing aqueous bioavailability (Tian et al., 2020; Alvarado et al., 2021). The sediment compartment, with Cu, Cr, Ni, Pb, and Zn consistently detected across all seasons and sites, functioned as an integrating sink that scavenged pollutants from the water column through sorption and settling, while also being subject to redox reactions and microbial transformations that influenced contaminant fate (Chiaia-Hernández et al., 2022).

The higher As concentrations in the water column during the rain-onset period are consistent with the region's geology. In alkaline lakes such as Chapala, As adsorbs strongly onto Fe and Mn oxyhydroxides at high pH, accumulating in sediments during the dry season. Rainfall runoff from the watershed, where volcanic tuffs are naturally enriched in As, likely remobilizes this geogenic As into the water column (de Anda et al., 2019).

However, the localized detection of multiple metals in interstitial water at CH6 during the warm-dry season (with Cd and Pb exceeding NOM-001-SEMARNAT-2021 limits) demonstrated that this sedimentary sink was not permanent, but rather locally susceptible to remobilization (SEMARNAT, 2022). This interstitial enrichment, consistent with the severe hypoxia (DO 18.06%) and negative ORP (−65.28 mV) recorded at that site during the warm-dry season, indicated a localized redox-driven process at the sediment-water interface rather than generalized water column contamination. Unlike CH7, which is located closer to the Zula River confluence and receives direct contaminant inputs, CH6 is situated farther from this confluence, where hydrodynamic disturbance is less intense; however, the reducing conditions at this site were sufficient to promote the reductive dissolution of metal oxides and the release of Cd and Pb into pore water (Ramírez-Sánchez and Fajardo-Montiel, 2024). This phenomenon is consistent with Ramírez-Sánchez and Fajardo-Montiel (2024), who documented similar metal mobilization under negative ORP conditions in Lake Chapala. Our findings further identified CH6 as a specific hotspot where localized redox conditions, rather than hydrodynamic disturbance, drive metal remobilization into interstitial water, a pattern previously undocumented in this lake.

4.4. Bioaccumulation patterns of pesticides and heavy metals in fish

Results showed clear differential bioaccumulation of pesticides and trace metals among fish species, which was linked directly to their ecological niches and was governed by species-specific factors influencing contaminant uptake, storage, and elimination (Nyeste et al., 2024; Alvarado et al., 2021).

4.4.1. Pesticide bioaccumulation

Benthic species (carp and Chapala catfish) exhibited the highest accumulations of glyphosate, AMPA, and endosulfan sulfate compared with pelagic feeders (tilapia and bass). This differential accumulation was governed by habitat and diet: as bottom feeders, carp and catfish were directly exposed to contaminated benthic organisms and resuspended sediment particles (Cornish et al., 2023; Bonansea et al., 2018). This pattern of higher accumulation in benthic species compared with pelagic feeders was consistent with the expected role of feeding ecology in governing contaminant uptake (Nyeste et al., 2024). Our Zn concentrations in carp (5.27-26.6 mg/kg) and catfish (1.0-11.67 mg/kg) were generally lower than those reported by Alvarado et al. (2021) for the same benthic species in Lake Chapala (carp: 8.84-16.05 mg/kg; catfish: 5.66-12.64 mg/kg), indicating reduced metal loading in recent years. These values are also comparable to those reported in other freshwater ecosystems; for instance, Yazman et al. (2025) documented a Zn concentration of 5.058 mg/kg in rainbow trout (*Oncorhynchus mykiss*) from Çamlıgöze Dam in Türkiye, while Ustaoğlu and Yüksel (2024) reported values ranging from 2.905 to 11.914 mg/kg in four fish species (*Carassius gibelio, Squalius cephalus, Esox lucius*, and *Tinca tinca*) from lagoon lakes in the Black Sea region, and Yüksel et al. (2025) documented concentrations between 4.386 and 5.854 mg/kg in three species (*Carassius gibelio, Squalius cephalus, and Esox lucius*) from Miliç Wetland. Notably, unlike many studies that frequently report the presence of multiple metals such as Cr, Ni, Pb, Cd, and As in fish tissues (Yazman et al., 2025; Yüksel et al., 2025; Ustaoğlu and Yüksel, 2024), zinc was the only metal consistently detected above the LOD in our samples; other metals remained at or below detection limits in fish muscle, suggesting a shift away from severe metallic pollution in Lake Chapala's biota during the study period.

Atrazine and metolachlor showed consistently low bioaccumulation potential, consistent with their relatively low log Kow values (~2.6 and ~3.05), limiting partitioning into lipid-rich tissues (Gianni et al., 2022; do Carmo et al., 2023).

The significant positive correlation observed between glyphosate and zinc concentrations in fish tissue (Spearman's ρ, $p < 0.05$), together with the pronounced dominance of WBF over SBF and the preferential accumulation in benthic feeders, documents a clear co-occurrence pattern. One possible interpretation of this pattern is that dissolved metals such as zinc may play a role in glyphosate uptake. It has been proposed that the formation of stable, soluble complexes between glyphosate/AMPA and divalent metal cations (Zn^{2+}, Ca^{2+}) in the water column could neutralize their charge, increase effective lipophilicity, and thereby facilitate membrane permeation (Mertens et al., 2018; Mohy-Ud-Din et al., 2023). However, this interpretation is speculative without speciation data or controlled exposures. The correlation could

equally arise from shared agricultural sources or seasonal transport dynamics. Thus, we treat the correlation as a descriptive finding and the metal-complex mechanism as a hypothesis requiring experimental testing. If confirmed, this hypothesis would be relevant to reported glyphosate-metal co-exposure effects in agricultural communities (Jayasumana et al., 2015; Wanigasuriya et al., 2011).

4.4.2. Heavy metal bioaccumulation

Zn was the only element consistently detected at quantifiable concentrations across all species and seasons, while the others remained at or below LOD. This pattern indicated a shift away from severe industrial metallic pollution, contrasting with mining-impacted basins where multiple metals exceeded permissible limits for human consumption (Affandi and Ishak, 2019). Although lake sediments act as reservoirs for various heavy metals (Ramírez-Sánchez and Fajardo-Montiel, 2024), only Zn accumulated consistently in fish tissue.

Our Zn concentrations in carp (5.27-26.6 mg/kg) and catfish (1.0-11.67 mg/kg) were generally lower than those reported by Alvarado et al. (2021) for the same benthic species in Lake Chapala (carp: 8.84-16.05 mg/kg; catfish: 5.66-12.64 mg/kg), indicating reduced metal loading in recent years. This pattern of higher accumulation in benthic species compared with pelagic feeders was consistent with the expected role of feeding ecology in governing contaminant uptake (Nyeste et al., 2024).

The invasive common carp showed Zn concentrations that were comparable to or exceeding those of the native Chapala catfish, while both substantially exceeded pelagic feeders, demonstrating that feeding ecology could override predictions based on invasion status alone (Franco et al., 2025).

4.4.3. Multivariate analysis of contaminants in fish

Fish weight showed minimal contribution to either axis, demonstrating that body mass did not determine contamination patterns, consistent with evidence that changes in bioaccumulation in aquatic food webs were mediated by species traits and trophic interactions rather than size (South et al., 2025). The differential accumulation between benthic and pelagic species reflected feeding ecology as a key functional trait, with implications for the nutritional quality and safety of fish available to local communities as invasive species such as carp altered food webs.

The elevated contaminant burden in the native Chapala catfish was of particular conservation concern, as this species had already been in decline due to habitat alteration and competition with invasive species (Juárez-Aguilar and López-Gómez, 2024). Chronic exposure to the reported levels of Zn, glyphosate, and AMPA reported here may have represented an additional stressor that impaired reproduction, immune function, or foraging efficiency (Ray and Shaju, 2023). The catfish thus served as a sentinel for cumulative chemical and biological stressors in the lake. The concurrent detection of elevated zinc and glyphosate/AMPA in these fish was consistent with an agricultural chemical signature, as zinc is a common additive in livestock feed and phosphate fertilizers that contributed to adjacent water bodies through runoff (Jayasumana et al., 2015; Mertens et al., 2018), and with studies that documented widespread occurrence of these herbicides in aquatic ecosystems globally (Bonansea et al., 2018).

4.5. Ecological and human health risk assessment

The safety of Lake Chapala's ecosystem services is of the utmost importance for human wellbeing, as these services are intricately linked to ecosystem health. Although HQ values for fish consumption had been below 1, suggesting negligible non-carcinogenic risk, this finding demanded cautious interpretation. The ecological risk assessment sounded an unequivocal alarm, with ΣRQ values reaching 936 during rain onset driven by diuron, DDD, and atrazine at concentrations acutely toxic to aquatic life. Although hazard quotients for fish consumption remained below 1, this should not be interpreted as a reassuring result. Lakeside communities are faced with multiple pathway exposure of multiple contaminants which is consistent with CKD diagnoses worldwide (García-García et al., 2020; Imbulana et al., 2020). The context of Lake Chapala and Poncitlan has similarities to epidemiological studies in regions with high rates of chronic kidney disease of unknown-origin (CKDu). In Sri Lanka's CKDu-endemic zone, for example, systematic water quality evaluations had consistently documented widespread fecal contamination in groundwater, with over 90% of wells showing total coliforms and half testing positive for *Escherichia coli*. These field studies may not always show a direct statistical correlation between coliform levels and CKDu prevalence, largely because fecal contamination was so pervasive that it became an environmental constant across affected regions. Nonetheless, populations experiencing high kidney disease rates were chronically exposed to microbiologically unsafe water. Importantly, this microbial contamination typically coincided with other potential renal stressors, such as elevated water hardness and magnesium concentrations (Imbulana et al., 2020). Furthermore, the risk characterization presented in this manuscript was based solely on oral exposure using national dietary guidelines that may underestimate localized consumption of fish which tends to be far greater in riparian communities (Tamale et al., 2017; He et al., 2022). Furthermore, our exposure assessment did not account for dermal, inhalation, or incidental ingestion pathways. These routes may have been especially significant in lakeside communities, where Smith et al. (2020) reported total coliforms in 81% of household water samples and 70% of tap water samples, and arsenic in 62% of tap water sources. Consequently, current risk estimates likely underestimated the total exposure burden. Future assessments would benefit from local consumption surveys and site-specific exposure data to refine these estimates and capture the full range of exposure pathways relevant to these communities.

In contrast, the ecological RQ values indicated acute risk during pulsed contamination events that occurred precisely when biological productivity was highest, potentially cascading upward to reduce prey availability and alter energy flow in ways that affected fish populations and the fisheries that supported local communities. The persistence of significant ecological concern across all seasons (ΣRQ 935.87-3.21) indicated this was not a seasonal anomaly but an enduring threat. This pattern mirrored findings in Laguna del Cisne (Uruguay), where DDT exhibited the highest ecological risk among all pesticides evaluated (RQ up to 230), driven by agricultural runoff during peak application seasons (Rodríguez-Bolaña et al., 2024). The ecological risk thus served as a precursor to human health risk, as a degraded ecosystem could channel contaminants more efficiently to top predators, including humans, through food web degradation (Carvajal-Quintero et al., 2026), including humans.

Hydrological drivers of precipitation and riverine input were clear drivers of contaminant dynamics in Lake Chapala. The rain onset season represented the highest risk period due to first runoff events transporting accumulated oxidizable substances from the watershed (Murillo-Delgado et al., 2021). The exclusive detection of DDT and its metabolites during rain onset season argued against steady release from legacy sedimentary deposits within the lake, instead indicating episodic runoff-driven inputs. Previous surveillance programs focused on conventional parameters in the water column were inherently inadequate to capture the spatially and temporally variable fluxes at sediment-water interfaces (Hansen and van Afferden, 2001), which underscored the necessity of expanding monitoring to include lotic inflow sites into lacustrine environments for prioritization in remediation and risk assessments.

These findings illustrate the relevance of the One Health framework, which recognizes that human, animal, and environmental health are inextricably linked. The multipathway exposure scenario we documented through contaminated water, sediment, and fish is consistent with emerging hypotheses regarding CKD in agricultural regions (García-García et al., 2020; Imbulana et al., 2020). Confirmation of

causal relationships between environmental contamination and human disease outcomes, however, will depend on dedicated epidemiological and toxicological investigations that fall outside the scope of the present study.

Universal access to safe drinking water was a priority, yet 78.6% of residents felt the government had not guaranteed this right (Smith et al., 2020). The documented presence of glyphosate (100%) and dimethoate (93%) in urine samples from affected lakeside communities confirmed that environmental contamination was translating directly into human body burden and that current regulatory approaches had failed to protect public health (Lozano-Kasten et al., 2021; Sierra-Diaz et al., 2019). Mexico's RENAMECA monitoring program must urgently expand its scope to include organic micropollutants. Public health campaigns should encourage consumption of pelagic species (tilapia, bass) over benthic species (carp, catfish) to reduce dietary contaminant intake while preserving vital protein access for vulnerable communities (Trasande et al., 2010).

5. Conclusions

This study provides the first integrated assessment of pesticide and metal contamination across water, sediment, and biota in Lake Chapala over a full hydrological cycle. The results demonstrate that seasonal hydrology critically drives contaminant dynamics, with first-flush rainfall remobilizing legacy DDT metabolites while the sediment-water interface facilitates cadmium and lead release at concentrations exceeding Mexican norms.

Among fish, benthic feeders accumulated significantly higher zinc burdens than pelagic species, indicating that feeding ecology rather than invasion status is the primary determinant of bioaccumulation. A correlational pattern between glyphosate and zinc in fish tissue was observed, though this relationship requires mechanistic validation.

The ecological risk assessment sounded an unequivocal alarm, with ΣRQ values reaching 936 during rain onset driven by diuron, DDD, and atrazine at concentrations acutely toxic to aquatic life. Although hazard quotients for fish consumption remained below 1, it could not be interpreted as reassurance, as lakeside communities faced multi-pathway, multi-contaminant exposure, a multiple-hit scenario consistent with CKD diagnoses worldwide.

The broader implications of this work were threefold. For research, our findings demanded non-targeted screening to capture the full chemical exposome, as well as rigorous epidemiological designs linking environmental data with human health outcomes. Ongoing work is testing whether Lake Chapala's contaminant mixture induces cytotoxicity, genotoxicity, and endocrine disruption in human renal cells. For policy, Mexico's monitoring framework needs to expand to include organic micropollutants and interstitial water at river confluences, and a glyphosate phase-out should be accelerated with post-ban monitoring. For communities, actionable guidance should include promoting consumption of pelagic fish (tilapia, bass) over benthic species and prioritizing universal access to safe drinking water. Collectively, these data established a critical baseline and provided actionable evidence to regulate glyphosate, diuron, and legacy organochlorines.

CRediT authorship contribution statement

Dania Marlene Ceballos-Vázquez: Writing – review & editing, Writing – original draft, Visualization, Investigation, Formal analysis, Data curation, Conceptualization. **John Nightingale:** Data curation, Formal analysis, Visualization. **Carlos Yebra-Montes:** Formal analysis, Data curation, Visualization. **Josie South:** Writing – review & editing, Validation, Supervision, Funding acquisition, Conceptualization. **Oriana Landa-Cansigno:** Writing – review & editing, Methodology. **Ofelia Yadira Lugo-Melchor:** Methodology. **Karina G. Coronado-Apodaca:** Writing – review & editing, Visualization, Supervision. **Hector Barrios-Pina:** Writing – review & editing, Supervision, Project administration, Funding acquisition, Visualization, Conceptualization. **Osiris Díaz-Torres:** Writing – review & editing, Writing – original draft, Visualization, Validation, Supervision, Project administration, Methodology, Investigation, Funding acquisition, Conceptualization.

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.

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Appendix A. Supplementary data

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Data availability

The dataset supporting this study is available on Zenodo under DOI 10.5281/zenodo.19503596.

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