



Topics of limnological research in Mexico

Coordinator
Alfredo Pérez Morales

UNIVERSIDAD DE COLIMA

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*This book is dedicated to
Dr. Singaraju Sri Subrahmanyam Sarma,
in gratitude for all his teachings in the world of limnology.*



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Utilization of Zooplankton in Environmental Risk Assessment in Mexico

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Abstract

This brief review focuses on the use of zooplankton as a key indicator to evaluate environmental risk in the aquatic ecosystems of Mexico. It explores some ecotoxicological studies that assess the impacts of toxic agents on individual and population levels. The role of zooplankton in food webs, susceptibility to environmental changes, and significance as an indicator of both direct and indirect pollution effects are emphasized. The study underscores the crucial role of ecotoxicological tests in preventing environmental damage, guiding decision-making, and managing ecosystems. Special attention is given to the ecotoxicology of rotifers, cladocerans, and copepods, providing insights into cultivation protocols, their relevance in ecotoxicology, and key species used in Mexico. It further explores testing methodologies, advantages, and the range of pollutants assessed. The review also examines the geographical distribution of zooplankton research in Mexico, underlining the necessity to broaden investigations to diverse aquatic systems. Challenges, such as the importance of including native species in toxicity studies and the development of specific protocols for freshwater copepods, are addressed.

Keywords

Ecotoxicology, Acute toxicity, Chronic toxicity, Rotifers, Cladocerans, Copepods.

Introduction

Environmental Risk Assessment

The environmental risk assessment (ERA) is a method that assesses the probability of adverse ecological effects arising from the exposure of organisms and communities to one or more chemical compounds, either currently happening or likely to occur (Di Lorenzo et al., 2023). In Mexico, researchers have incorporated the use of aquatic invertebrates into ERA for several years. The selection of these organisms in these evaluations is based mainly on ecological relevance (validity), reliability (reproducibility), representative test species, and sensitivity, as noted by Breitholtz et al. (2006). ERA is carried out through ecotoxicological studies conducted in both laboratory and field settings. Ecotoxicology is comprised of three disciplines: ecology, toxicology, and chemistry and examines the effects of toxic compounds at different levels of biological organization, including individuals, population, and community levels (Pastorino et al., 2024). Measurable outcomes encompass physiological homeostasis, reproductive behavior, morphological alterations, and mortality (Zimmermann and Sures, 2023). This approach employs sensitive organisms as reliable indicators, connecting compound exposure to organism response. These data enable eco-toxicologists to identify concentrations indicating adverse conditions (OECD, 2011).

Ecotoxicological studies play a crucial role in both preventing environmental damage and understanding the chemical characteristics of the environment. They are indispensable in decision-making processes related to resource protection and ecosystem management (Relyea & Hoverman, 2006). These assessments provide valuable insights into the effects of pollutants on aquatic organisms and ecosystems, thereby influencing the formulation guiding the development of environmental protection guidelines. Additionally, these studies help identify sensitive stress indicators, facilitating in the evaluation of mitigation measures (Montalvo & Luque, 2009).

Mexico has a wide variety of aquatic species that can be used in ecotoxicological tests. These tests are essential for conducting a comprehensive Environmental Risk Assessment (ERA), particularly studies focused on evaluating water quality over time and space in waterbodies impacted by pollutants (Santos-Medrano et al., 2007; Guzmán-Colis et al., 2011). In the present review, we emphasize the importance of ecotoxicological studies carried out in Mexico that use native zooplankton species as model organisms. We have compiled various relevant research that addresses the use of rotifers, cladocerans, and copepods in the assessment of environmental risk associated with wastewater treatment plant effluents, heavy metals, pesticides, cyanotoxins, pharmaceuticals, and microplastics, all of which are frequent pollutants in the aquatic environments of Mexico and other regions of the world.

Zooplankton Applications in Ecosystem Health

Zooplankton is vital for aquatic ecosystems, contributing to food webs, nutrient cycling, biogeochemical processes, and algal bloom regulation (Declerck & de Senerpont, 2023). Marine zooplankton, spanning 12 phyla, is diverse, with copepods being the most abundant, constituting 80 % of biomass (Gasca, 2010; Peijnenburg & Goetze, 2013). In freshwater communities, rotifers, cladocerans, and copepods dominate, with rotifers significantly contributing to biomass (Sarma & Nandini, 2017; Elías-Gutiérrez & Ortiz, 2017).

Zooplankton is highly vulnerable to environmental changes, impacting other organisms through trophic interactions (Gutiérrez & Gagneten, 2011). Due to its ease of cultivation and sensitivity, with well-known nutritional requirements and short life cycles, it's an ideal model for ecotoxicological evaluations (Declerck & de Senerpont, 2023). Ecotoxicological tests use rotifers, cladocerans, and copepods as indicators to assess direct and indirect effects (Elías-Gutiérrez & Gagneten, 2011). For example, effluent quality is often assessed based on physical and chemical parameters alone; however, zooplankton have proven useful in assessing the effectiveness of wastewater treatment plants through acute toxicity tests (Torres-Guzmán et al., 2010).

Zooplankton Species Used Globally

Researchers employ zooplankton models tested in controlled conditions or micro/mesocosms (Moreno et al., 2022). Microcosms maintain consistent but less complex conditions, while mesocosms, offering greater complexity, simulate controlled or natural environments indoors or outdoors (Lozano, 2020). Outdoor systems, while authentic, face weather unpredictability, complicating replication and causing data variation. Researchers must align conditions with their study focus and logistical factors, especially in ecotoxicology (Hjorth et al., 2006).

According to De Meester et al. (2023), the majority of zooplankton tests have centered around the *Daphnia* genus due to its well-researched ecology over the past 120 years. Copepod research has focused on risk assessment in both marine and freshwater environments, particularly emphasizing the naupliar stage (López, 2018). *Acartia tonsa* is common in bioassays for evaluating environmental hazards due to its ecological significance and adaptability (Rotolo et al., 2021). Studies indicate that *Tisbe battagliai*, *Tigriopus japonicus*, *Nitocra spinipes*, and *Mesocyclops leuckarti* are more sensitive to various contaminants than *Daphnia magna* (OECD, 2011).

Rotifers are cost-effective tools for environmental protection. The *Brachionus* genus (*B. plicatilis*, *B. calyciflorus*, and *B. havanaensis*) is recognized by the American Public Health Association for assessing xenobiotics, endocrine disruptors, and disinfectants in both freshwater and marine environments (Alayo & Iannaccone, 2002; Sarma et al., 2014).

Zooplankton Groups in Mexico for Environmental Risk Assessment

As of January 2024, in the Scopus database, there are roughly 450 research studies on zooplankton in Mexico, with 80 of them specifically focusing on environmental risk assessment. These studies have highlighted 50 different species of zooplankton with the primary attention given to three main groups: rotifers, cladocerans, and copepods. Other groups, including Anostraca (*Artemia franciscana*), Amphipoda (*Hyalella azteca*), and Ostracoda (*Diaphanocypris meridana*), have also been studied (Fig.1). The majority of research has been conducted in entities such as the State of Mexico, Mexico City, Aguascalientes, Veracruz, Baja California Sur, and Quintana Roo. However, there are still areas, like Oaxaca, Durango, Zacatecas, Colima, and Coahuila, where research is scarce. This emphasizes the need to promote research in these areas to gain a better understanding of the environmental risk in Mexico.

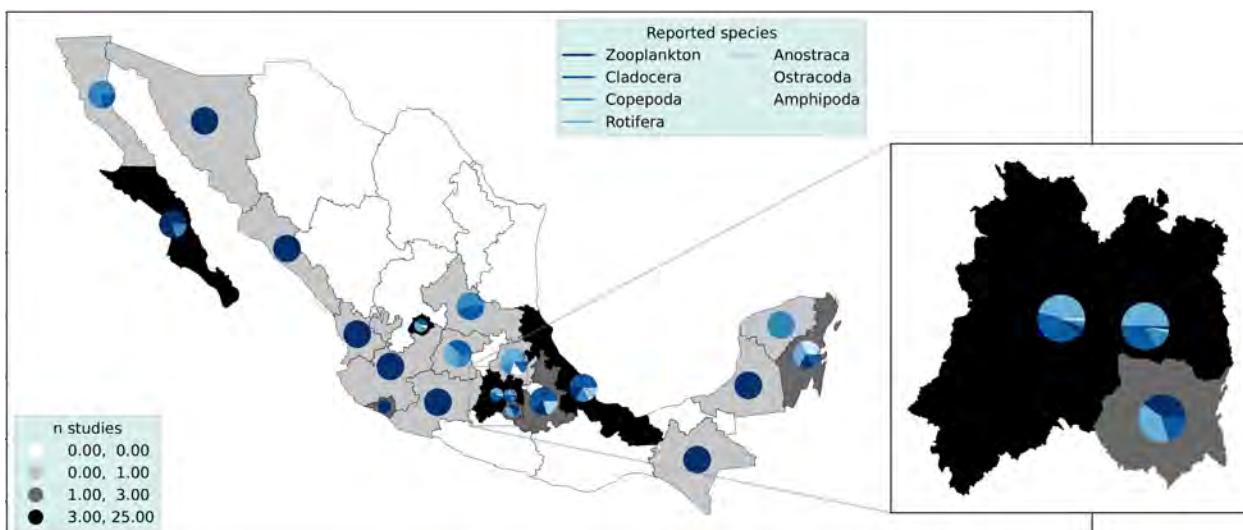


Figure 1. Distribution of Research on Zooplankton in Mexico with a Focus on Environmental Risk and the Main Groups Studied.

Rotifers

Diversity, Behavior, and Reproductive Strategies

Rotifers are microorganisms (approximately 50–2,000 µm in length) ubiquitously found in diverse ecosystems, ranging from fresh and brackish waters to marine environments worldwide. Phylum Rotifera has two classes: Pararotatoria with subclass Seisonidea and Eurotatoria with subclasses Bdelloidea and Monogononta (Fontaneto & Plewka, 2021). Globally, approximately 2,300 species have been described, while in Mexico, around 400 species have been recorded (Sarma et al., 2021). In general terms, the life cycle of mo-

nogonont rotifers begins with the hatching of amictic females from diapause eggs. When environmental conditions are conducive, such as optimal temperature, salinity levels and food availability, reproduction occurs asexually through parthenogenesis.

Conversely, in unfavorable situations, the reproductive process takes a sexual phase. In the sexual phase, mictic females produce diapause eggs stored in the sediments of aquatic systems and can remain viable for many years (Fontaneto & Plewka, 2021). The distinctive ability of rotifers to produce diapause eggs makes them valuable resources for laboratory experiments, including ecotoxicological studies (Won et al., 2017).

Use of Rotifers in Ecotoxicology

Early ecotoxicological studies involving rotifers commenced in the 1980s, as reported by Halbach et al. (1983). Since then, rotifers have been pivotal in evaluating environmental risks linked to a spectrum of emerging contaminants, including pharmaceuticals, pesticides, and microplastics. Additionally, they have contributed significantly to assessing heavy metals, emerging pollutants, and the impact of harmful algal blooms. Various reasons support the position of rotifers as model organisms in this ecotoxicology: a) their ease of culture and manipulation in the laboratory; b) a short life cycle (7-20 days), which facilitates short-term analysis of demographic parameters such as average lifespan, life expectancy, reproductive rates, generation time and increase rates; c) parthenogenetic reproduction, which ensures genetic homogeneity and rapid population growth; d) its high sensitivity to changes in water quality; and e) ecological relevance and reliability in reproducibility (Dahms et al., 2011; Rico-Martínez et al., 2017; Won et al., 2017). These characteristics meet the requirements to consider rotifers as representative test species in environmental risk assessments.

Main Rotifer Species in the Ecotoxicology of Mexico

Recently, Sarma et al. (2021) carried out an exhaustive compilation of rotifer species present in freshwater bodies in Mexico. Among the rotifer families analyzed it was observed that the most diverse were Flosculariidae (50 species), Brachionidae (51 species), Leucanidae (68 species), Notommatidae (48 species), and Trichocercidae (31 species). It is relevant to highlight that the most significant diversity of species was found in the state of Mexico, where 323 species were recorded, followed by Michoacán with 164, Veracruz with 155, Aguascalientes with 150, and Yucatan with 129 species reported. In Mexico, various species of rotifers serve as model organisms in ecotoxicological tests. According to the data included in the Scopus database (November 2024), rotifer species used in ecotoxicological studies in Mexico include *Anuraeopsis fissa*, *Asplanchna sieboldii*, *Brachionus calyciflorus*, *B. angularis*, *B. rubens*, *B. havanaensis*, *Lecane hamata*, *L. luna*, *L. quadridentata*, *L. papuana*, *Euchlanis dilatata*, and *Plationus patulus*.

The basic rotifer bioassays include acute toxicity tests, like the 24 h LC₅₀ (lethal concentration for 50 % of the population) and the EC₅₀, indicating the effective concentration inhibiting 50 % of biological responses to contaminants. Chronic toxicity tests assess life table parameters and population growth in organisms exposed to toxicants (Dahms et al., 2011).

In Mexican aquatic ecosystems, *B. calyciflorus* is commonly used in studies on environmental risks. For example, Zamora-Barrios et al. (2017) evaluated the effects of crude extracts of cyanobacteria detected in Lake Nabor Carrillo, which is part of what was once Lake Texcoco (Mexico City), on *B. calyciflorus* isolated from the same waterbody. Thus, demonstrating the impact that cyanobacteria blooms in tropical waters have on key species. The rotifer *B. angularis*, isolated from a pond in the Park of Tezozomoc (Mexico City), has been helpful in evaluating the effects of methyl parathion, a commonly used insecticide in Mexico, to eradicate insect pests (Gama-Flores et al., 2004). This study illustrates the potential risk associated with the presence of pesticides in aquatic systems and their impact on the population dynamics of invertebrates, which contend with constant changes in water quality and food availability. It also underscores the organisms' usefulness in short-term assessments of environmentally relevant pesticides in Mexico, particularly in agricultural areas. In Aguascalientes, Tovar-Aguilar et al. (2019) used the rotifer *L. pa-puana* to evaluate the effects of the pharmaceutical diclofenac, detected in surface and groundwater due to its extensive use in human and veterinary health. This study exhibits the vulnerability of zooplankton to relevant emerging pollutants and stresses the potential for bioaccumulation of xenobiotics, which can induce disturbances in the ecological structure of aquatic environments. Heavy metal pollution in aquatic environments is a growing concern due to high concentrations, persistence, and biomagnification. The first reports of lead biomagnification in predatory rotifers were published by Rubio-Franchini and Rico-Martínez (2008, 2011) at Niagara Dam in Aguascalientes. Their research focused on the species *A. brightwellii* and emphasized the importance of conducting *in situ* studies while confirming the findings through laboratory tests. Similarly, another study highlights the relevance of assessing the toxicity of metals (Al, Fe, and Zn) in the San Pedro River using acute toxicity tests on *L. quadridentata* (Torres-Guzmán et al., 2010). This approach allowed the estimation of each metal's contribution to overall toxicity, identifying zinc as the most toxic metal, underscoring the value of combining field and laboratory analyses for a comprehensive evaluation of metal pollution.

A recent study in Manatí Lagoon, a protected area in Cancún (Quintana Roo), evaluated the risk through zooplankton, including rotifers (Demidof et al., 2022). These key indicators of aquatic health underscore the urgency of addressing pollution in protected environments. This comprehensive approach, involving the evaluation of key organisms such as rotifers, highlights the urgent need for effective strategies to counteract the de-

trimental effects of heavy metal pollution in aquatic environments, especially in protected areas such as Laguna Manatí. The rotifers *P. patulus* and *A. sieboldii*, isolated from Lake Xochimilco (Mexico City), were used to analyze the effects of microplastics and their interaction with heavy metals on the predator-prey dynamics and demographic variables of the rotifers (Hernández-Lucero et al., 2023). This study revealed that mixtures of heavy metals and microplastics can accentuate the vulnerability of key species to pollution. Thus, it highlights the importance of rotifers in the ecological risk assessment of microplastics in Mexico, emphasizing the need to understand the interaction between these contaminants and aquatic fauna to preserve the health of ecosystems.

Challenges

Most ecotoxicological studies with freshwater rotifers focus on the cities of Mexico and Aguascalientes. This is presumed due to the presence of groups of experts on the subject in these states. While there is evidence of ongoing efforts in the states of Veracruz and Yucatan. It's crucial to intensify these efforts and expand research to diverse aquatic systems in Mexico, leveraging the potential of rotifers. These organisms can be tools in developing mitigation strategies and deepening our understanding of the effects of pollution in different aquatic environments in the country. The study of the effects of heavy metals on rotifers is more common than research on other contaminants. It's essential to explore the environmental risks of emerging contaminants of global relevance, such as pharmaceutical waste and microplastics which still require more detailed investigations. Furthermore, it is crucial to consider more realistic scenarios in studies that reflect aquatic systems contaminated by a mixture of substances, an evaluation made possible thanks to the advantages that rotifers offer as model organisms. Surprisingly, in rotifer studies in Mexico, environmental genomics still needs to be fully integrated despite being a tool that could provide a deeper understanding of the toxicity mechanisms of various toxic substances.

Cladocerans

Diversity, Behavior, and Reproductive Strategies

Cladocerans, commonly referred to as “water fleas,” belong to the Phylum Arthropoda and the class Branchiopoda, encompassing four recognized Orders: Anomopoda, Ctenopoda, Haplopoda, and Onychopoda. With a size range of 0.25 to 18 mm, there are nearly 850 described species, including 150 recorded in Mexico (Cervantes-Martínez et al., 2023). They are the connection between the microbial loop, primary producers, and higher-level consumers (Kalinowska, 2015).

Cladocerans employ their thoracic appendages to filter particles, optimizing nutrient consumption for efficient growth and reproduction (Riisgård, 2015; Smirnov, 2017). Most

species exhibit facultative parthenogenesis, where males are associated with seasonality or environmental unpredictability. They produce resting eggs (ephyppia), serving as a genetic reservoir and facilitating passive dispersal (Bernatowicz et al., 2018). However, certain species within the *Daphnia pulex* complex may display obligatory parthenogenesis (Huynh et al., 2023).

Use of Cladocerans in Ecotoxicology

The utilization of cladocerans in bioassays is supported by their easy maintenance in laboratory settings and sensitivity (Terekhova et al., 2018). Since the 1970s, environmental agencies have standardized protocols for evaluating diverse toxicants using cladocerans (Versteeg et al., 1997). Toxicological studies on cladocerans involve acute tests (LC₅₀ determination within 24-48 h) (OECD, 2004; U.S. EPA, 2002) and chronic tests, assessing life history, physiology, and molecular responses over extended periods (>30 % of the life cycle) (Connors et al., 2022). Population growth and life table experiments offer vital insights into their fitness, stress responses, and reactions to chemical compounds (Sibly & Hone, 2002; Wilson et al., 2006; Sarma & Nandini, 2006).

Main Cladocerans Species in the Ecotoxicology of Mexico

In Mexico, 17.6 % of the globally recognized cladoceran species have been identified, with ongoing efforts to expand this number. However, by 2008, only 1 % of watersheds had been thoroughly explored, indicating vast potential for further study (Elías-Gutiérrez et al., 2008a). Additionally, the use of molecular markers has brought to light a large number of cryptic species (Elías-Gutiérrez et al., 2008b).

The first study to use cladocerans as a model assay in Mexico was focused on evaluating the chronic toxicity of wastewater from the paper industry (Martínez-Jerónimo et al., 1993). Since then, there has been an exponential increase in publications exploring the effects of various chemical, physical, and biological compounds, such as heavy metals, pesticides, surfactants, personal care products, pharmaceuticals, hormones, phytotoxins, microplastics, and wastewater. These studies have been conducted across a limited range of species, including *Daphnia magna*, *D. pulex*, *D. laevis*, *D. exilis*, *D. schoedleri*, *D. ambigua*, *Ceriodaphnia dubia*, *Moina macrocopa*, *M. micrura*, *Alona glabra*, *Diaphanosoma birgei*, *Bosmina longirostris*, and *Macrothrix triserialis*. This surge in research highlights the growing recognition of the important role of cladocerans in aquatic ecotoxicology within the Mexican scientific community.

Cladocerans have proven to be a reliable model organism in Mexico's field of environmental assessment. A pioneering study conducted by López-López and Serna-Hernández (1991), linked seasonal zooplankton variation in reservoirs and identified species like *Bosmina longirostris*, *Diaphanosoma birgei*, and *Daphnia parvula* as excellent indicators of

water quality changes associated with eutrophication (Mendoza-Chávez et al., 2022). Recent research in San Luis Potosí revealed a correlation between arsenic levels and microcrustacean diversity in reservoirs, suggesting *Simocephalus punctatus* as a potential reliable bioindicator. Furthermore, saprobity indices have been devised for assessing water quality in the Xochimilco canals (Nandini et al., 2016).

Studies on urban lakes in Mexico City have focused on the impact of different feeding conditions, particularly in the presence of toxin-producing cyanobacteria. Cladocerans, including *Simocephalus mixtus*, *Daphnia mendotae*, *D. pulex*, *Moina micrura*, *M. macrocopa*, and *Ceriodaphnia dubia*, actively consume cyanobacterial cells, leading to alterations in filtration rates and fitness (Pineda-Mendoza et al., 2012; Pérez-Morales et al., 2014, 2020; Nandini et al., 2020) or inducing cyanotoxin production (Pérez-Morales et al., 2015). Additionally, cladocerans have been observed to accumulate heavy metals, cyanotoxins, and microplastics, subsequently transferring them within the food chain (Rubio-Franchini et al., 2016; Zamora-Barrios et al., 2019; Manríquez-Guzmán et al., 2023).

Research with Cladocera strains isolated from Mexican waterbodies examined the impact of Hexavalent Chromium on *Ceriodaphnia dubia*, emphasizing precise short-term assays, including volume, exposure duration, and temperature measurements (Martínez-Jerónimo & Martínez-Jerónimo, 2023). In Aguascalientes, a study revealed high susceptibility of the indigenous species *Alona guttata* to pesticides, even at concentrations similar to guava field applications, causing chronic exposure-related somatic growth alterations. Furthermore, the Holarctic species *Daphnia magna* has been utilized to evaluate the impact of toxic substances such as nonsteroidal anti-inflammatory drugs (NSAIDs) on oxidative stress and genetic material damage (González-González et al., 2014). Additionally, Mexican laboratories participated validating a two-generational reproduction test (Barata et al., 2017). Recent researches are focused on the effects of multiple stressors to assess the synergistic or antagonistic impacts of a wide range of emerging contaminants found in domestic, textile, and hospital wastewater, as well as pesticides. These contaminants include a mixture of detergents, metals such as Zinc, Cadmium, and Arsenic, anti-corrosion agents, cardioactive drugs, antibiotics, NSAIDs, antidepressants, etc. (Hernández-Zamora & Martínez-Jerónimo, 2019; Aguilar-Aguilar et al., 2023; Hernández-Zamora et al., 2023). All the works above highlight the relevance of cladocerans as valuable tools for understanding the response of aquatic ecosystems to factors such as pollutants, changes in feeding, and environmental conditions, providing crucial information for the management and conservation of aquatic resources in Mexico.

Challenges

Mexican legislation designates *Daphnia magna* as a sentinel for toxicity tests (NMX-AA-087-SCFI-2010). However, smaller herbivores such as, *Diaphanosoma*, *Moina*, *Ceriodaphnia*, *Chydorus*, *Alona*, and *Macrothrix* genus, dominate Mexican freshwater ecosystems. These species have shorter lifespans, faster reproduction, lower fecundity, and potentially greater sensitivity than *D. magna*. Martínez-Jerónimo et al. (2008) have proposed to replace *D. magna* with the American cladoceran *Daphnia exilis* in ecotoxicological bioassays due to its taxonomic similarities. Following the same approach, Santos-Medrano and Rico-Martínez (2019) suggest determining the relative sensitivity of native species to *D. magna* using the formula proposed by Von der Ohe and Liess in 2004:

$$S = \log \left(\frac{LC_{50} \text{ of } Daphnia magna}{LC_{50} \text{ of } i} \right)$$

Where: S =relative sensitivity; $LC_{50} \text{ of } Daphnia magna$ = LC_{50} value for *D. magna*, and LC_{50i} =experimental LC_{50} for a species i . A zero value indicates a sensitivity equal to that of *D. magna*, a positive value suggests that *D. magna* is less sensitive, and a negative value indicates that *D. magna* is more sensitive.

In a preliminary experiment designed to assess the impact of Potassium dichromate, *Daphnia magna* exhibited LC_{50} values of 0.93 at 24 h and 0.66 mg L⁻¹ at 48 h. The native species *C. dubia*, *D. laevis*, and *S. vetulus* showed greater sensitivity. *D. laevis* presented an LC_{50} of 0.34 mg L⁻¹ at 48 h, resulting in an S value of 0.28. *S. vetulus* was the most sensitive species, with a $S=0.81$ (Fig.2).

Despite the absence of *D. magna* records in Mexican water bodies, recent studies identify it in Ciénegas del Lerma (Espinoza-Rodríguez, 2023). This underscores the importance of considering native species in ecotoxicological studies for a more realistic scenario and reducing the likelihood of introducing exotic species.

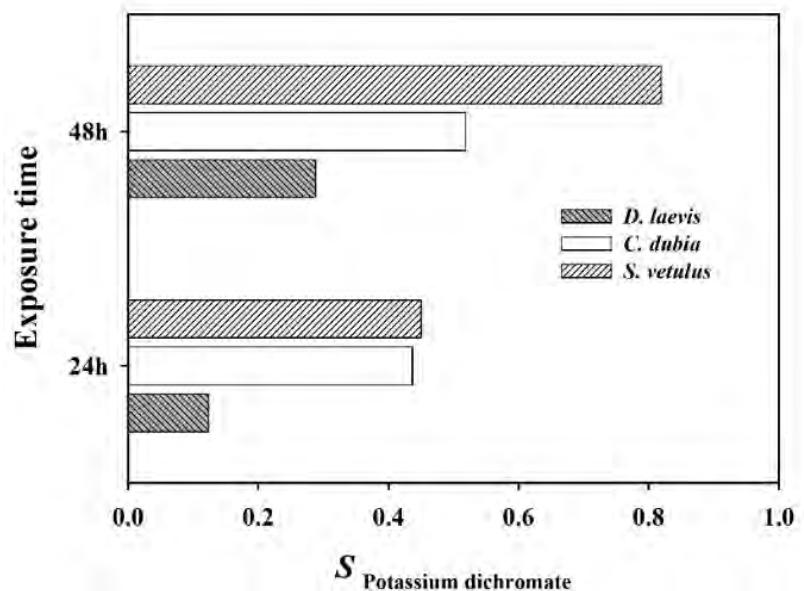


Figure 2. Relative Sensitivity Values of Native Species (*Ceriodaphnia dubia*, *Daphnia laevis*, and *Simocephalus vetulus*) Exposed to the Reference Toxicant (Potassium dichromate) during 24 and 48 h Experiments.

Copepods

Diversity, Behavior, and Reproductive Strategies

In general, most copepods are found in marine or brackish waters (~14,000 registered), with around 3,000 species in freshwater (Uc-Castillo et al., 2022). Copepods in continental waters are classified into three orders: Calanoida, Cyclopoida, and Harpacticoida (Dole-Olivier et al., 2000). Approximately 110 species have been documented in Mexican aquatic ecosystems (Gómez & Morales-Serna, 2014). Copepods typically measure between 1 and 5 mm in length, exhibiting a cylindrical body, segmented exoskeleton, and articulated appendages for swimming and feeding. Copepods display sexual dimorphism, and sexual reproduction is the most common form of reproduction; however, parthenogenesis has been observed in certain harpacticoid species (Poulin, 1996). Most copepods hatch from fertilized eggs, involving the union of a spermatophore, delivered by the male, to the copulatory pore of the female (Reid & Strayer, 1994). Copepod development involves eleven stages, including six naupliar and five copepodite stages with molting occurring between each stage and metamorphosis from the last naupliar to the first copepodite stage (Kwok et al., 2015).

Use of Copepods in Ecotoxicology

Copepods are acknowledged as highly effective bioindicators of ecosystem pollution, with various toxicity testing protocols developed, including acute tests, multi-generation life cycle

tests, and short-term toxicity tests, assessing responses in individual actions, immune and endocrine processes, development, growth, and reproduction (Hussain et al., 2020). Since the 1940's, copepods have remained a popular model organism with most toxicity studies utilizing either static or static-renewal systems. Among copepod species, *Amphiascus tenuiremis*, *Nitocra spinipes*, and *Acartia tonsa* have seen the establishment of standardized full life-cycle testing protocols (Raisuddin et al., 2007). Unfortunately, freshwater copepods have not received sufficient attention, lacking protocols for toxicity tests. Nevertheless, *Mesocyclops* genera have been identified as a suitable option (Kulkarni et al., 2013).

Main Copepods Species in the Ecotoxicology of Mexico

Research on copepods in Mexico has traditionally focused on their geographic distribution and taxonomic description. However, there is a growing interest and recognition of the important role copepods play in ecotoxicological studies. Several copepod species have become essential models for evaluating environmental impacts and conducting ecotoxicological tests. A simple search in the Scopus and Web of Science databases using the words "toxicology," "copepods," and "environmental assessment" shows that some of the marine species examined include *Acartia tonsa*, *A. clausi*, *A. spinata*, *A. lilljeborgii*, *Corycaeus amazonicus*, *Temora discaudata*, *Subeucalanus subcrassus*, *Acrocalanus longicornis*, *Calanus pacificus*, *Euterpina acutifrons*, and *Pseudodiaptomus euryhalinus*. Additionally, freshwater species such as *Acanthocyclops robustus*, *A. vernalis*, *A. americanus*, *Mastigodiaptomus montezumae*, *Paracyclops novenarius*, and *Eucyclops chihuahuensis* have also been used.

The research on copepods in Mexico is limited but existing studies emphasize their importance. A specific study examined the reproductive response of the copepod *A. clausi* to the toxic dinoflagellate *Gymnodinium catenatum* suggesting that this species plays a crucial role in controlling red tides in Concepción Bay (Palomares-García et al., 2006). Another work identified the high tolerance of *P. novenarius* in a water body with high Arsenic concentrations ($>50 \text{ mg L}^{-1}$), revealing that despite the high concentrations, there was no impact on its morphology or development (Uc-Castillo et al., 2022). Additionally, sensitivity and response were evaluated using oxidative stress biomarkers on *A. americanus* exposed to Cadmium, Chromium, Copper, Mercury, Manganese, Nickel, and Lead, emphasizing the need to understand how copepods, both marine and freshwater, respond to various environmental factors and contaminants (Sobrino-Figueroa et al., 2020). A recent investigation examines the impact of invasive species, such as *M. pehpeiensis*, not only from an ecotoxicological perspective but also considering their effects on biodiversity reduction and the potential impact on Mexican planktonic communities (Valencia-Vargas et al., 2023).

Challenges

Ecotoxicological risk assessment commonly involves selecting species based on their sensitivity to various toxins and their suitability for laboratory cultivation. However, this approach often overlooks the diverse life history strategies copepods adopt in their natural environments. This oversight could have substantial implications for these species' vulnerability to contaminant exposure. Consequently, there is an urgent call for detailed and carefully orchestrated research within the realm of copepod ecotoxicology. Such research is pivotal in addressing the current disparity between ecological and ecotoxicological studies on copepods, with the goal of accurately identifying and incorporating representative species into assessments.

Conclusions and Recommendations

The focus of this comprehensive brief review is on the significance of ecotoxicology in Mexican aquatic ecosystems, specifically directing attention towards zooplankton. The urgency to advance research in this field highlights the need to explore diverse ecosystems, particularly those in understudied regions. The inclusion of native species in toxicity studies emerges as a crucial aspect to ensure realistic outcomes and prevent the introduction of invasive species. Key approaches proposed for enhancing understanding include conducting detailed research on emerging contaminants (pesticides, animal pharmaceuticals, pharmaceuticals and personal care products, industrial compounds), heavy metals, detergents, as well as integrating environmental genomics into zooplankton studies. Furthermore, the establishment of standardized protocols, specifically with freshwater copepod species, is emphasized as an imperative need. Overall, these future perspectives collectively aim to strengthen the knowledge base in aquatic ecotoxicology in Mexico, providing valuable insights for the enduring preservation of the country's aquatic ecosystems.

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Authors' Contributions

CAZB, UAR, MGRM, JMEH, and FJTM designed, analyzed, and wrote the study; JMEH and CAZB developed the graphic representation. FJTM acquired financial funds.

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This book takes a significant step in showcasing the relevance of limnology to our survival. Freshwater habitats, though they cover less than 1 % of the Earth's surface, are home to a substantial portion of the world's biodiversity—at least 10 % of all known species. Freshwater habitats and the biodiversity they support are under threat. Moreover, our survival depends on access to high-quality freshwater. This book not only highlights the beauty of limnology and the scientific methods used to study it, but it also draws attention to the major causes of biodiversity loss in freshwater ecosystems. It shows all readers what it means to deal with inland waters as a scientist interested in understanding ecosystems and protecting them.

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