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Chapter- 2

Life in the Balance: Zooplankton's Battle in a Changing Environment

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Abstract:

Zooplankton are often overlooked but are vital components of marine and freshwater ecosystems. Zooplankton are pivotal in nutrient cycling and ecosystem dynamics as they transfer energy between primary producers and higher trophic levels. However, unprecedented growth in human population and industrialization have exposed aquatic environments to various pollutants, threatening zooplankton communities worldwide. Nutrient over-enrichment, primarily from sewage discharge and agricultural runoff, has caused eutrophication in water bodies. It is altering species composition and favouring the proliferation of certain zooplankton groups while decimating others. As a byproduct of industrialization, heavy metals have infiltrated aquatic ecosystems, accumulating in zooplankton and propagating up the food chain. It poses grave risks to human and ecosystem health. Microplastics (MPs) infiltrating aquatic environments also threaten zooplankton, impairing feeding, growth, and reproduction and altering gene expression. The emergence of pharmaceuticals and antibiotics as environmental contaminants further compounds the plight of zooplankton, disrupting reproduction, survival, and ecological resilience. Pesticides, pervasive in agricultural runoff, harm zooplankton communities significantly, jeopardizing ecosystem stability. Climate change compounds the problem in zooplankton communities by inducing range shifts and phenological changes, altering community dynamics, and heightening vulnerability to other stressors. Regular monitoring of zooplankton has emerged as an invaluable indicator of ecosystem function. As researchers strive to unravel the complex interplay of stressors reshaping aquatic ecosystems, the status of zooplankton communities can signal the urgent need for concerted conservation efforts and proactive management strategies to safeguard the ecological balance of our aquatic realms.

Introduction:

Aquatic ecosystems worldwide are under severe threat of degradation due to various anthropogenic activities (Roy et al., 2022; Das et al., 2023). The need to maintain the ever-expanding human is releasing hazardous chemicals and modifying the landscapes at an

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unprecedented rate. There is a significant focus on assessing how multiple stressors caused by human-environment interactions and climate change are altering marine and freshwater ecosystems. There is a growing interest among scientists, policymakers, governments, and stakeholders in assessing the health of species, populations, communities, and ecosystems as a whole (Patra and Madhu, 2009; Dutta et al., 2014; Mallick and Panigrahi, 2018; Biswas et al., 2023).

Zooplankton are unsung heroes of marine and freshwater food web. Zooplankton is composed of a diverse array of species with wide-ranging sizes. These animals create an energy transfer link between primary producers and higher trophic levels. Many years of research have shown that zooplankton are sensitive to minute environmental changes. In aquatic ecosystems, anthropogenic factors such as heavy metals, microplastics (MPs), pesticides, antibiotics, and pharmaceuticals significantly affect zooplankton abundance, diversity, distribution, reproduction, and development. In some studies, there is evidence of anthropogenic stressors on zooplankton gene regulatory pathways as well. The changes in zooplankton communities can also act as bioindicators of ecological stress. Zooplankton groups like rotifers, cladocerans, copepods, etc., show different responses toward different stressors. On one hand, human activities and eutrophication can promote the growth of certain types of zooplankton. On the other hand, these same factors can cause increased mortality in other species. Along with creating an adverse effect on the overall health of zooplankton, bioaccumulation of pollutants also passes the harmful chemicals to higher trophic levels. In this article, we have discussed the complex nature of the influence of several anthropogenic stressors and their effects on zooplankton communities.

Role of zooplankton in the ecosystem:

Zooplankton are the key component of any aquatic ecosystem and act as an important trophic link between producers and higher trophic levels to transfer energy and matter. Grazing of the zooplankton also serves to cycle nutrients and carbon from the microbial loop. Marine zooplankton contribute to several ecosystem services. The biggest contribution is in ecosystem-supporting services - nutrient cycling, food sources to higher trophic levels, larval recruitment to fisheries, and a refuge for various other organisms. They play a crucial role in regulating nitrogen and phosphorus cycling and controlling their availability to phytoplankton. By consuming organic nitrogen and releasing dissolved organic nitrogen (DON) through their excreta, these organisms play a crucial role in supporting heterotrophic bacterial growth and exerting control on primary production. Zooplankton also transports particulate organic nitrogen (PON) to depth via the production of faecal pallets (Steinberg & Saba, 2008)**.** Essential provisioning services include wild food and the production of fish meals. Meta zooplankton is utilized as a sustainable alternative to traditional fish feed in aquaculture, serving as a live food supplement. Certain biomedical applications are also included, especially from jellyfish. Zooplankton is involved in carbon sequestration in the deep sea by sinking faecal pellets and sedimentation of dead zooplankton. It is essential for climate regulating services. For a detailed discussion about zooplankton's contribution to ecosystem services, we encourage readers to refer to recent review articles on this topic (Botterell et al., 2023). In the context of environmental pollution by human activities, zooplankton can also be of disservice in certain instances. It is a well-established fact that a diverse range of pollutants, heavy metals included, are biomagnified through zooplankton, according to research studies. This highlights the importance of being vigilant about the disposal and management of such pollutants to prevent further harm to our environment and its inhabitants. In the following sections, we discuss the interaction of some classes of pollutants with zooplankton communities. Studies from field observations and laboratory studies are included.

Effect of nutrient over-enrichment on zooplankton communities:

Nutrient over-enrichment in natural aquatic ecosystems by sewage discharge and agricultural and industrial runoff has become a major matter of concern. The primary impact of nutrient overload is eutrophication. The excess nutrient content (especially nitrogen and phosphorus) boosts the growth of phytoplankton and algal biomass, which causes oxygen depletion and dead zone formation in the water body. These incidents may convert a top-down controlled ecosystem into bottom-up control (Fernández-Alías et al., 2022). Nutrient contamination and eutrophication decrease species richness and favor the growth of more specialized and dominant zooplankton. Eutrophication promotes the growth of cyanobacterial blooms. Cyanobacteria are considered a poor-quality food source for most zooplankton, including copepods and cladocerans, because they lack many essential lipids. Its anti-grazing traits like toxicity, size, and low nutritional value reduce the fitness of zooplankton-grazers such as *Daphnia sp.* and enhance the mortality rate (Ger et al., 2016). Contamination of water by total nitrogen, total ammonia nitrogen, nitrite, and nitrate is one of the main stressors for the zooplankton community. However, studies found that different zooplankton groups show specific responses towards specific contamination. Due to high tolerance, the diversity and abundance of rotifers (*Brachionus rotundiformis* and *Brachionus rubens)* are directly proportional to the total ammonia nitrogen content. Whereas diversity and abundance of cladocera (*Moina* sp.) and copepods (*Acartia* sp.) are inversely proportional to total ammonia nitrogen content (Yang et al., 2017). Another threat caused by nutrient contamination is the entry of harmful chemicals into the food web through zooplankton. One example is the transfer of polychlorinated biphenyls (PCB) to the upper trophic level by plankton grazer *Daphnia pulicaria* (Lynn et al., 2007). Furthermore, harmful nutrient overload may affect zooplankton's lifespan and reproductive health.

Impact of metal toxicity in zooplankton communities:

With increasing industrialization in developed and developing countries, metals are increasingly released into aquatic ecosystems. Industrial and domestic sewage effluents, electronic waste, mining and oil drilling operations, etc., are major sources of heavy metal pollution in water. Zooplankton can uptake heavy metals directly from water or via metals accumulated in phytoplankton. As they are the food for higher trophic levels, accumulated nonbiodegradable metals in the zooplankton community play a significant role in transferring toxic metals to fish and humans, leading to public health concerns. Even though some metals are essential micronutrients (i.e., Cu, Zn, Cr, Mo), higher accumulation of these can cause significant physiological problems in animals. Nonessential heavy metals, such as Pb, Hg, and Cd, are established toxins affecting numerous biological activities in animals. Thus, the trophic transfer of these metals remains a very active area of research.

Heavy metals such as Cd, Pb, Cu, Fe, Cr, As, and Zn have a significant detrimental effect on the zooplankton by abundance, population, growth, body size, egg production, and egg hatching. Industrialization near coasts has increased the discharge of heavy metals in coastal, estuarine waters. Studies from the Bay of Bengal (BoB) indicate the presence of multiple heavy metals (Ni, Cu, Zn, Pb, Fe, Mg, Co, and Cr) which have major genotoxicity in zooplankton (Thirunavukkarasu et al., 2020). In the southwest part of the Bay of Bengal, the order of metal accumulation in zooplankton was found to be in the order - Fe > Zn > Mn > Cr > Ni > Pb > Cu > Ce > La > Co > U > Cd. A strong positive correlation was found between bioaccumulation in mesozooplankton and soluble metals in water in case in case of Zn and Cr (Achary et al., 2020). At the western Bay of Bengal, heavy metal accumulation study in copepods reveals that inshore communities have much higher bioaccumulated heavy metals compared to offshore samples. Higher Pb, Cd, and Ni levels in inshore zooplankton are a grave concern. Presence of strong East India Coastal Currents in the western BoB can potentially transport the copepods with accumulated heavy metals along the Indian coastline. Thus, contaminating food webs of distant places with heavy metals (Singaram et al., 2023).

One study from China indicated that urbanization causes both nutrient and heavy metal pollution in adjacent aquatic ecosystems. Higher density and abundance of some heavy metal-resistant species of zooplankton, such as, *Synchaeta oblonga* may serve as an indicator of polluted waters near highly urbanized areas. Other species like *Keratella cochlearis* and *Anuraeopsis fissa* can indicate slightly polluted water in weakly urbanized environments (Shen et al., 2021).

Zooplankton and microplastics – junk food for the primary consumers of aquatic ecosystems:

We are living in an “age of plastics,” and plastic has emerged as a ubiquitous threat to aquatic environments. In recent years, there has been an increased emphasis on the study of microplastics, which are tiny plastic particles with a diameter of less than 5mm, and their impact on the environment and human health. aquatic creatures can consume microplastics in a few ways. They may eat MPs that resemble their natural food, consume prey that has already consumed MPs, or ingested MPs while filtering feeding. Additionally, benthic creatures can take in MPs during sediment mixing. Evidence of MPs affecting different aspects of zooplankton life has emerged from laboratory and field observations involving marine and freshwater species. The chief concern is the transfer of bioaccumulated MPs in zooplankton to higher trophic levels, and if MPs impact the zooplankton community, then the potential disruption of fish communities that depend on them. MPs can affect zooplankton's feeding activity, growth, development, excretion, increased mortality, and reproduction. Higher MP concentrations in water lead to a greater intake of MPs in zooplankton (Messinetti et al., 2018). The size of MPs overlaps with the size of phytoplankton that zooplankton graze on. MPs can obstruct feeding and damage the digestive system, reducing food intake (Cole et al., 2013). The disruption of food intake affects the growth, development, and reproduction processes in a cascade. Polystyrene microbeads lead to reduced fecundity in the copepod *Tigriopus japonicas* due to failure to develop egg sacs (Lee et al., 2013). Also, in another copepod *Calanus helgolandicus*, even if egg production is not disrupted, they are smaller in size and fail to hatch (Cole, Lindeque, Fileman, Halsband, & Galloway, 2015). It is concerning that MP accumulation is detected in Arctic and Antarctic zooplankton, indicating their potential entry into the pelagic food web in those regions (Wilkie et al., 2023). The ageing of microplastics can promote greater ingestion by marine zooplankton, as shown by studies with copepods *Calanus finmarchicus* and *Acartia longiremis*. This may be due to the coating of aged microplastics with biofilms (Vroomet al., 2017). In the aquatic environment, the coating of microplastics with algal-derived infochemical dimethyl sulphide (DMS) can promote increased uptake and bioavailability of MPs in zooplankton (Botterell et al., 2020).

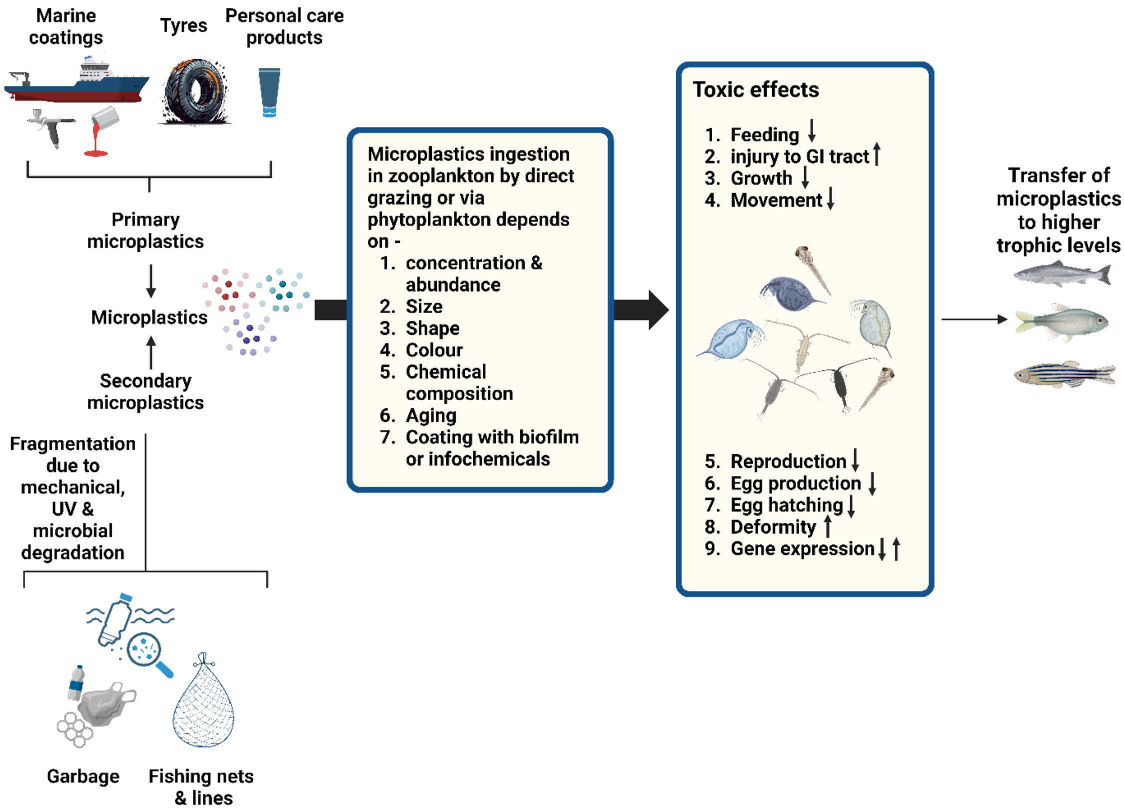

Figure 1. Negative impact of microplastics on zooplankton – Uptake of microplastic in zooplankton depends on several factors. It harms the zooplankton community via different toxic effects. Ingested microplastics can be transferred to higher trophic levels through the zooplankton. Image created with BioRender.com

At the molecular level, ethylene acrylic acid copolymer particles have been shown to alter expression levels of genes of central metabolism, oxidative stress, ovulation, and moulting in *Daphnia magna* (Coady et al., 2020). Polystyrene microbead exposure can change the oxidative stress pathways by modulating thioredoxin reductase (TRxR) and arginine kinase (AK) genes (Tang et al., 2019). Microplastics can be converted into nanoplastics in the environment, and they are potentially even more bioavailable. A recent report of rotifers converting microplastics into nanoplastics has raised concerns about the contribution of zooplankton to aquatic nanoplastic pollution (Zhao et al., 2023). A summary of impact of microplastics on zooplankton is represented in figure 1.

Antibiotics and pharmaceuticals as newly emerging pollutants affecting zooplankton:

Anthropogenic activities have released various emerging concerns (CECs) into the environment. Among them, a growing body of work indicates the presence of antibiotics and other pharmaceuticals in aquatic ecosystems. The primary sources of antibiotic contamination in aquatic ecosystems are from human and veterinary use, administration in aquaculture, and intense animal farming. Their concentrations are high enough to cause adverse effects on the resident zooplankton. Analyses suggest that bioaccumulation and biomagnification in the planktonic food web are possible for contaminant antibiotics such as tetracycline, oxytetracycline, roxithromycin, lomefloxacin, ofloxacin, etc (Tang et al., 2020). Antibiotics can also cause dysbiosis of gut microbiota in zooplankton. Many researchers have used Daphnia magna as a model organism to study the effect of antibiotics. In these zooplankton, tetracycline has been demonstrated to diminish reproduction and abundance. In the absence of adequate food, the toxicity of this antibiotic is increased (Akbar et al., 2020). Due to norfloxacin exposure, heartbeat rate and feeding efficiency were decreased in *D. magna*. It increased the time ratio of vertical to horizontal swimming (TVH) and the duration of quiescence (Pan et al., 2017). Lomefloxacin is also recorded to cause oxidative stress-induced cellular damage in these animals (Luo et al., 2018). Sometimes, multiple antibiotics are detected in aquatic ecosystems. To model such scenarios, Daphnia magna was exposed to a cocktail of antibiotics (aztreonam, erythromycin, and sulfamethoxazole), which decreased the associated microbiome diversity (Cooper, Tjards, Rischling, Nguyen, & Cressler, 2022). There are reports of other antibiotics and pharmaceuticals hampering density, reproduction, and survivorship in rotifers as well (González-Pérez et al., 2016; Wang et al., 2017). This is a newly emerging field of study, and further research is necessary to unravel the complex interactions antibiotics can have on zooplankton.

Disruption of zooplankton communities by pesticides:

Pesticides have become widespread contaminants in aquatic ecosystems. Wetlands close to agricultural fields are worst affected due to agricultural discharge, runoff, and drift of pesticide sprays. Scientists relied on laboratory-based studies utilizing model organisms (i.e., *Daphnia* sp.) to study the LC50 values and life history, physiological and behavioural effects.

Worryingly, relatively newer pesticides, such as imidacloprid, which is favoured for low toxicity in vertebrates and short environmental persistence, have been shown to disrupt zooplankton in water bodies. Feeding activity is significantly reduced in *Daphnia magna* even at sub-lethal concentrations of imidacloprid. This may be due to abnormal motility to avoid the substance and higher metabolic costs to detoxify it (Pestana et al., 2010). Zooplankton such as cladocerans, copepods, and rotifers produce egg banks as dormant stages to overcome adverse environmental conditions. They are deposited in the sediments and stay there till hatching under favourable conditions. The application of glyphosate-based pesticides has been documented to impair emergence from egg banks and reduce zooplankton diversity (Gutierrez et al., 2017).

Currently, a complex mixture of insecticides is present in affected water bodies, and researchers are focusing more on community-level disruptions. One study found 29 different pesticides in a lake associated with declining abundance of metazoan zooplankton. Chlorpyrifos and cypermethrin were the main culprits identified to cause this decline (Kong et al., 2022). Long-term studies indicate that insecticide mixtures can continue to impact natural systems for several weeks, even after they are no longer detectable in water (Hasenbein et al., 2016).

An exciting area of research is the emergence of pesticide resistance in zooplankton. Pesticide resistance is of grave concern when it is found in the target pest species. However, research has shown that if a population of zooplankton in a community is resistant to pesticides, then it helps the community to be resilient towards that contaminant. One study showed that if a *Daphnia pulex* population resistant to AChE-inhibiting insecticide chlorpyrifos is present in an aquatic community, it may help to maintain the community dynamics even when exposed to other similarly acting insecticides (i.e., malathion, carbaryl). Mesocosms with insecticide-sensitive *D. pulex* populations experience phytoplankton blooms after exposure to insecticides. This was avoided if resistant *D. pulex* populations were present. If sodium channel-inhibiting insecticides (i.e., permethrin, cypermethrin) are added, it leads to a reduction in the abundance of both chlorpyrifos-sensitive and resistant *D. pulex* populations (Hasenbein et al., 2016). Future studies should focus on insecticide-resistant zooplankton populations in natural freshwater and estuarine ecosystems.

Climate change and zooplankton – web of complex interactions:

Over the past decade, accelerated climate change has threatened to drastically alter the aquatic system's environmental parameters, which greatly impacts zooplankton. Climate change imparts changes in marine ecosystems through different mechanisms. The structure of the zooplankton community is influenced by the warming of the upper layer of the ocean, which affects the process of nutrient enrichment and water column stratification. Under well-mixed cold water conditions, the surface layers are supplied with nutrients that favor the population of large copepods. But in warm stratified waters, nutrient supply to surface layers is hampered. This favors the zooplankton community being dominated by jelly fishes, ctenophores, salps, etc. (Richardson, 2008). Range shift is documented in calanoid copepods of North Atlantic Ocean. They are shifting northward at a rate of 23.16 km/yr due to rising sea surface temperatures (Gregory et al., 2009). It is important to note that these shifts are not consistently observed, and they vary significantly in strength and direction, often being specific to a particular species. A consequence of warming ocean temperature is attributed to the advancement of zooplankton phenology. Global warming is causing earlier peak zooplankton abundance mainly due to the advancement of spring. For instance, the biomass of *Neocalanus plumchrus*, copepod found in Subarctic Pacific Ocean, is peaking 73 days earlier per 1℃ rise in temperature (Ratnarajah et al., 2023). Ocean acidification is another great concern. Even though copepods were thought to be resilient to ocean acidification, new research has suggested that the nauplii stages of their life cycle suffer higher mortality due to acidification (Cripps et al., 2014). Copepod *Centropages velificatus*, when exposed to simulated thermal stress of heatwave, were found to have higher mortality and reduced egg production. This thermal stress also made it more susceptible to anthropogenic stressors such as oil spills (Hernández Ruiz et al., 2021). Freshwater zooplankton are also not spared from the ill effects of climate change. Frequent and increasingly intense heat waves are a clear sign of climate change. When phytoplankton are cultured under heatwave conditions, they cannot nourish freshwater zooplankton (Kim et al., 2024). The influence of climate change on zooplankton dynamics is a multifaceted phenomenon. Scientists are engaged in documenting the changes already happening in zooplankton communities and trying to model future changes.

Conclusion:

Zooplankton communities in marine and freshwater ecosystems face diverse anthropogenic stressors, which are only increasing in intensity. Globally, evidence of considerable changes in abundance, distribution, and physiological and behavioural alterations in zooplankton is being reported. Scientists are constantly striving to study the effect of environmentally relevant concentrations of toxicants on zooplankton dynamics. The situation is even more complex as zooplankton in a particular place are exposed to various stressors belonging to different categories. What impact these assemblages of stressors have on zooplankton is slowly being revealed. Zooplankton is reported to accumulate both heavy metals and organochlorine pesticides, simultaneously providing a pathway for the movement of these toxicants to other organisms (Basu et al., 2021). Warming waters can influence the effect of other toxicants. Increased temperatures can prolong the long-term adverse effects of pesticides in *Daphnia* sp. (Knillmann, Stampfli, Noskov, Beketov, & Liess, 2013). The presence of pharmaceutical contaminants can enhance the derogatory impact of climate change in freshwater ecosystems (Duchet et al., 2024). Climate change can alter the exposure of marine species to microplastics by disrupting their reproductive cycles and behaviours (Haque & Fan, 2023).

Monitoring every single species in an ecosystem is practically impossible. As a result, it is beneficial to develop zooplankton bioindicators that can be used to assess the status and trends within ecosystems (Burger, 2006). Zooplankton plays a crucial role in the ecosystem, yet it is rarely utilized commercially. This unique characteristic, combined with the fact that it can reflect the impact of various environmental stressors, makes it an ideal candidate for the role of a bioindicator. Consistently monitoring zooplankton is extremely valuable for planning and evaluating the outcomes of conservation efforts.

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