Content available at: https://www.ipinnovative.com/open-access-journals

IP International Journal of Medical Microbiology and Tropical Diseases

Journal homepage: https://www.ijmmtd.org/

Original Research Article

Effect of heavy metals on *paramecium* found in different water bodies of Bhiwandi

Hiba Mohd Shujaat Shaikh[1,*], Mukesh Pimpliskar[2]

[1] *Molecular Pathlabs LLP, Thane, Maharashtra, India*
[2] *Dept. of Biotechnology, K.M.E Society's G. M. Momin Women's College, Bhiwandi, Maharashtra, India*

ARTICLE INFO

Article history:
Received 28-03-2022
Accepted 23-04-2022
Available online 07-06-2022

Keywords:
Heavy metals
Ferrous Sulphate
Mercuric Chloride
Zinc Chloride
acute toxicity
Paramecium
ciliates
biological indicator

ABSTRACT

Paramecium is a unicellular organism, widely distributed in the freshwater environment, where heavy metals are common contaminants. *Parameciums* are very abundant group in aquatic ecosystems, which makes them effective biological indicators of water pollutants. The aim of this study was to determine the ability of *paramecium* to survive in various concentrations of three different heavy metals: Ferrous Sulphate ($FeSO_4$), Mercuric Chloride ($HgCl_2$) and Zinc Chloride ($ZnCl_2$). The ciliates were incubated in solutions with 0.3 to 1.5 ppm of $FeSO_4$, 0.01 to 0.05ppm of $HgCl_2$ and 0.10 to 0.95 ppm of $ZnCl_2$. at room temperature. Using compound microscope, microscopic observations of cell divisions rate was conducted after 24, 48, 72 and 120 hours of incubation in the tested solutions and was compared to the control sample. Microscopic observations revealed that all the concentrations of $HgCl_2$ were lethal for all samples except sample no. 3 while $FeSO_4$ and $ZnCl_2$ were found to be growth promoter for *paramecium* of all 10 samples.

1. Introduction

Water is one of the most important compounds in the ecosystem. Better quality of water is described by its physical, chemical and biological characteristics. The quality of water is a vital concern for mankind because it is directly linked to human health. Water sources were polluted by domestic wastage in rural areas whereas industrial wastages discharged into natural sources in urban areas.[1] Humans need water every day for various domestic, irrigation drinking purpose. When there was no revolution in industry and agriculture, water quality was near about good, but due to industrial and agricultural revolutions water which is collected in the various water resources gets highly polluted in various ways.

Heavy metals are considered very important and highly toxic pollutants. Eco-toxicologists and environmental scientists use the term heavy metals to refer to metals that have caused environmental problems. Heavy metals including both essential and non-essential elements have a particular significance in ecotoxicology, since they are highly persistent and all have the potential to be toxic to living organisms. Several heavy metals have been studied extensively over the last few decades. Inputs of these toxic heavy metals to the environment as a result of anthropogenic activities is difficult to measure due to the very large natural inputs from the erosion of rocks, wind-blowing dusts, volcanic activity and forest fires. Atmospheric and river inputs, dredging spoil, direct discharges, industrial dumping and sewage sludge are some of the important contributors to metal pollution, which lead to the release of toxic heavy metals to the marine environment. Acid rain resulting from dissolved hydrogen sulphide, sulphur dioxide and oxides of nitrogen has contributed to alterations of soil and freshwater acidity. As a consequence, there is an increase in the bioavailability of many heavy metals

* *Corresponding author.*
E-mail address: hibamsshaikh4160@gmail.com (H. M. S. Shaikh).

https://doi.org/10.18231/j.ijmmtd.2022.026

to freshwater biota. Metals are separated into the essential and non-essential in classes A and B, and in a borderline class. Class A metals: Calcium (Ca), Magnesium (Mg), Manganese (Mn), Potassium (K), Sodium (Na), Strontium (Sr) Class B metals: Cadmium (Cd), Copper (Cu), Mercury (Hg), Silver (Ag) Borderline metals: Zinc (Zn), Lead (Pb), Iron (Fe), Chromium (Cr), Cobalt (Co) Nickel (Ni), Arsenic (As), Vanadium (V), Tin (Sn). The world-wide emissions of metals into the atmosphere (thousands of tons per year) by natural sources is estimated as: Ni: 26, Pb: 19, Cu: 19, As: 7.8, Zn: 4, Cd: 1.0, Se: 0.4, (x103tons.Year-1), whereas, from anthropogenic sources: Pb: 450, Zn: 320, Ni: 47, Cu: 56, As: 24, Cd: 7.5, Se: 1.1 (x103 tons. Year-1). It is obvious from these numbers that Pb, Zn, Ni and Cu are the most important metal pollutants from human activities. Heavy metal pollution of freshwater has been shown to be extensive. The behaviour of metals in natural waters is a function of the substrate sediment composition, the suspended sediment composition, and the water chemistry. During their transport, the heavy metals undergo numerous changes in their speciation due to dissolution, precipitation, sorption and. phenomena which affect their behavior and bioavailability. Hence, heavy metals are sensitive indicators for monitoring changes in the aquatic environment. If aquatic resources are not properly managed and aquatic ecosystems deteriorate, then human health and well-being may be compromised.[2]

In the last decades, governments have promulgated strict regulations to guarantee the high quality of drinking water. Some metals are very harmful to freshwater organisms, such as cadmium or lead. On the other hand, metals like iron, copper, zinc or nickel, though they are essential components of living cells, at certain concentrations can be dangerous.[3] Aquatic environments include both fractions, soluble and inparticulate matter.[4] The way in which heavy metals affect the microorganisms is not clear. Scientists suggest metals block some enzyme systems or can interfere with some important cellular metabolites of protozoa and bacteria.[5] Source of Copper pollution is from metal painting, plating and mineral leaching into waste water. It is an essential trace element not very toxic to animals, toxic to plant and algae at moderate levels and harmful to human beings at high levels. Copper sulphate is extremely harmful to fishes. Even at low concentration of application, this may create toxic effects to trouts and fishes in acidic soft water. With increase in the water hardness, its toxicity to fish gets decreased. As compared to young fishes, the eggs of fishes get destroyed by the copper toxicity. Mercury is a widespread environmental toxicant and pollutant which induces severe alterations in the body tissues and causes a wide range of adverse health effects and also has adverse effects on animals, plants and microorganisms.[6]

The study of toxic and trace metals in the environment is more important in comparison to other pollutants due to their non-bio-degradable nature, accumulative properties and long biological half-lives. It is difficult to remove them completely from the environment once they enter into it. With the increased use of a wide variety of metals in industries and in our daily life, there is now a greater awareness of toxic metal pollution of the environment. Many of these metals tend to remain in the ecosystem and eventually move from one compartment to the other within the food chain. These essentially need the quantitative analysis of major pollutants specifically the heavy metals in water. Very scant data on these aspects is available from Thane district, especially areas in and around Bhiwandi, hence Bhiwandi city in Thane district, Western Maharashtra was selected as location for study.

Paramecium is a unicellular organism, belonging to ciliates, which play a significant role in aquatic ecosystems.[7] Ciliates are considered to be cosmopolitan organisms. They can be easily found in various types of watercourses and bodies of stagnant water. Ciliates mostly fed on bacteria and they are prey of organisms of higher taxons as well. That is why they represent important links in the trophic chains, where they mediate the flow both of biological substances and energy from a lower level to a higher one.[8] Additionally, they play an essential role in the purification processes of both aerobic and anaerobic biological wastewater treatment systems. Their sensitivity to environmental changes suggested their use as biological indicators of water pollution.[9]

Present research work aims at the analysis of the different water reservoirs of Bhiwandi and determines the effect of heavy metals on *Paramecium* found in different water bodies. Special focus of this research work is on the determination of the effects of Ferrous, Mercury, Copper and Zinc on the growth of *Paramecium* using the compound microscope in the water samples collected from the different water bodies of Bhiwandi.

2. Materials and Methods

In this study, *Paramecium* isolated from 10 water sample collected from different parts of Bhiwandi city, Dist. Thane, Western Maharashtra, India (Table 1). *Paramecium* cells were cultured in Hay-Wheat medium, at room temperature. The single cells were picked from the sample by micropipette and placed in a culture tube. Each tube included five cells of the studied organism. We can see various cell reactions after addition of various concentrations of different heavy metals and the concentrations were found from the research papers based on Bhiwandi and nearby area.[10,11]

The ciliates were incubated in 2ml Hay medium containing 100μl solutions of 0.30 to 1.5 ppm of $FeSO_4$, 0.01 to 0.05ppm of $HgCl_2$ and 0.10 to 0.95 ppm of $ZnCl_2$. To determine the toxicity of these three compounds, microscopic observations of the rate of cell division of

Table 1: Site collection area and type of water samples in Bhiwandi city

Sample No.	Area	Sample Type
1.	Samad nagar	Textile effluent
2.	Varal devi Talav	Pond water
3.	Khadipar	Well water
4.	Tandel mohalla	Well water
5.	Pokhran	Well water
6.	Eid'gah near Memon masjid	Well water
7.	Eid'gah near Muslim kabrastan	Well water
8.	Eid'gah near	Well water
9.	Eid'gah near Shiya masjid	Well water
10.	Waja mohalla	Well water

Table 2: Heavy metals used and their concentrations

Sample No.	Heavy Metals	Concentration (ppm)
1.	Ferrous Sulphate ($FeSO_4$)	0.30, 0.90, 1.50
2.	Mercuric Chloride ($HgCl_2$)	0.01, 0.03, 0.05
3.	Zinc Chloride ($ZnCl_2$)	0.10, 0.50, 0.95

Paramecia were carried out after 24, 72 and 120 hours of incubation in three different concentrations under room temperature (Table 2). Each sample was performed the same way and included the same number of cells.

3. Results

In this study, using compound microscope, carried out microscopic observations of the rate of cell division of paramecia after 24, 72 and 120 hours of incubation in different concentrations, at RT. According toTable 2, we can see various cell reactions after addition of various concentrations of different heavy metals and the concentrations were found from the research papers based on Bhiwandi and nearby area.[10,11] In case of Ferrous (FeSO4), occurrence of the reaction depended on the concentration of Ferrous. We used different concentrations of $FeSO_4$ from 0.3 to 1.5 ppm and a control sample. The reaction we were observing was the rate of cell divisions. It can be easily noticed that Ferrous was acting as growth promoter, in all the concentrations cells were dividing similarly to the control sample (Table 3). Mercury (HgCl2) is known as one of the most effective growth inhibitory/immobilizing agent for protozoa. In this study cell growth retardation was depended on mercury irrespective to the concentrations i.e. in all the samples except sample 3, death occurred in all the concentrations (Table 4). While no growth was observed after addition of Zinc ($ZnCl_2$), zinc was also acting as growth inhibitor for *paramecium*. (Table 5).

4. Discussion

The accumulation of the heavy metals in wastewater depends on many local factors, such as the type of industries in the region, lifestyle and awareness of the impact on the environment through the careless disposal of wastes. Being a city with extensive industrialization and civilization, water pollution by metal ions has emerged as one of the serious challenges currently faced by water service authorities in Bhiwandi. Hence, this study focused on the effect of heavy metal concentrations on *paramecium* isolated from 10 water samples collected from different water bodies of Bhiwandi city, Maharashtra.[12] Shunmugam, A.P et al. did the experiments in which they found the presence of heavy metals impacts on movement of Paramecia's, showing a significant decrease in their speed even when zinc chloride and copper sulfate concentrations are only half of those considered unsafe for drinking water.

Some metals like ferrous, mercury and zinc which were used in this study, though they are components of cells, at certain concentrations, turn out to be harmful. In this study, there were no lethal effect of Ferrous observed in fact they acted as *paramecium* growth promoter but all the concentrations of Mercuric Chloride i.e. 0.01 to 0.05ppm was found lethal for *paramecium* other than sample 3 because no change in number of cells was observed, and all the concentrations of Zinc was found to be lethal for all 10 samples.

5. Conclusion

The outcomes of the study revealed that the all 10 water samples of Bhiwandi, Maharashtra contain good population of *paramecium. Paramecium* can serve as a bioindicator to evaluate the quality index of effluents released from textile or leather industries, or, effluents rich in Ferrous, Mercury and Zinc contents. The studied concentrations of Ferrous served as growth promoter in all samples. Thus, as per the current analysis, it can be proposed that, Mercury and Zinc are toxic to freshwater ciliate *Paramecium*, but with the

Table 3: Effects of different concentration of ferrous Sulphate on *Paramecium* within 120 hours from the beginning of incubation of microscopic observation.

S. No.	Initial number of cells	Concentration of Ferrous 0.3 ppm				0.9 ppm				1.5 ppm			
		24h	48h	72h	120h	24h	48h	72h	120h	24h	48h	72h	120h
1.	5	10	18	35	60	6	15	32	67	5	7	13	22
2.	5	7	20	40	64	6	13	26	50	6	10	18	37
3.	5	12	27	48	75	10	23	45	82	8	20	43	73
4.	5	12	20	45	92	10	17	41	75	6	15	38	70
5.	5	10	25	52	103	7	19	47	91	5	9	25	52
6.	5	5	6	18	47	5	5	15	32	5	5	11	20
7.	5	10	21	47	79	7	17	40	74	6	15	35	67
8.	5	13	30	58	100	7	15	43	81	7	15	39	70
9.	5	11	28	62	116	8	20	49	90	5	12	30	66
10.	5	9	20	47	69	6	19	42	70	5	9	25	57

Table 4: Effects of different concentration of Mercuric Chloride on *Paramecium* within 120 hours from the beginning of incubation of microscopic observation.

S.No.	Initial number of cells	Concentration of Mercury 0.01 ppm				0.03 ppm				0.05 ppm			
		24h	48h	72h	120h	24h	48h	72h	120h	24h	48h	72h	120h
1.	5	0	0	0	0	0	0	0	0	0	0	0	0
2.	5	0	0	0	0	0	0	0	0	0	0	0	0
3.	5	5	5	5	5	5	5	5	5	5	5	5	5
4.	5	0	0	0	0	0	0	0	0	0	0	0	0
5.	5	0	0	0	0	0	0	0	0	0	0	0	0
6.	5	0	0	0	0	0	0	0	0	0	0	0	0
7.	5	0	0	0	0	0	0	0	0	0	0	0	0
8.	5	0	0	0	0	0	0	0	0	0	0	0	0
9.	5	0	0	0	0	0	0	0	0	0	0	0	0
10.	5	0	0	0	0	0	0	0	0	0	0	0	0

Table 5: Effects of different concentration of Zinc Sulphate on cells of *Paramecium* within 120 hours from the beginning of incubation of microscopic observation.

S.No.	Initial number of cells	Concentration of Zinc 0.10 ppm				0.50 ppm				0.95 ppm			
		24h	48h	72h	120h	24h	48h	72h	120h	24h	48h	72h	120h
1.	5	0	0	0	0	0	0	0	0	0	0	0	0
2.	5	0	0	0	0	0	0	0	0	0	0	0	0
3.	5	0	0	0	0	0	0	0	0	0	0	0	0
4.	5	0	0	0	0	0	0	0	0	0	**0**	0	0
5.	5	0	0	0	0	0	0	0	0	0	0	0	0
6.	5	0	0	0	0	0	0	0	0	0	0	0	0
7.	5	0	0	0	0	0	0	0	0	0	0	0	0
8.	5	0	0	0	0	0	0	0	0	0	0	0	0
9.	5	0	0	0	0	0	0	0	0	0	0	0	0
10.	5	0	0	0	0	0	0	0	0	0	0	0	0

studied concentrations of Mercury, maintenance of number of viable cells of *Paramecium* from sample 3 implies that these could be a mutant spp., as mercury is a known growth inhibitor for *Paramecium* at these concentrations.

6. Conflict of Interest

The authors declare no relevant conflicts of interest.

7. Source of Funding

None.

References

1. Sayyed JA, Bhosle AB. The study of zinc metal concentration by spectrophotometric method from Godavari river at Nanded. *Der Chemicasinica*. 2010;1(2):104–9.
2. Pravin S, Manisha T, Ravindra M. Sediment heavy metal contaminants in Vasai creek of mumbai: pollution impacts. *Am J Chem*. 2012;2(3):171–80.
3. Fraile A, Penche S, Gonzales F, Blazques ML, Munoz JA, Ballestera A, et al. Biosorption of copper, zinc, cadmium and nickel by Chlorella vulgaris. *Chemistry And Ecology*. 2005;21(1):61–75. doi:10.1080/02757540512331334933.
4. Violante A, Krishnamurti GSR, Pigna M. Mobility of Trace Elements in Soil Environments. In: Violante A, Huang P, Gadd G, editors. Biophysico-Chemical Processes of Metals and Metalloids in Soil Environments. Hoboken, NJ: John Wiley & Sons; 2008. p. 169–213.
5. Morgan GB, Lackey JB. BOD determinations in wastes containing chelated copper or chromium. *Sewage Industrial Wastes*. 1958;30(3):283–6.
6. Al-Bairuty GA, Boyle D, Henry TB, Handy RD. Sublethal effects of copper sulphate compared to copper nanoparticles in rainbow trout (Oncorhynchus mykiss) at low pH: Physiology and metal accumulation. *Aquat Toxicol*. 2016;174:188–98. doi:10.1016/j.aquatox.2016.02.006.
7. Lynn D. The Ciliated Protozoa: Characterization, Classification, and Guide to the Literature. 3rd Edm. Netherlands: Springer; 2008. doi:10.1007/978-1-4020-8239-9.
8. Cairns J, Gruber D. A comparison of methods and instrumentation of biological early warning systems1. *J Am Water Resour Assoc*. 1980;16(2):261–6. doi:10.1111/j.1752-1688.1980.tb02388.x.
9. Nicolau A, Manuel M, Nelson L. Physiological responses of Tetrahymena pyriformis to copper, zinc, cycloheximide and Triton X-100. *FEMS Microbiol Ecol*. 1999;30(3):209–16.
10. Choudhari KS, Chavan RP, Patil CL, Shinde DN. To determine the heavy metals with respect to chemical parameters of Bhiwandi (Maharashtra). *Int J Chem Stud*. 2016;4(2):42–5.
11. Jadhav AM, Singare PU. Studies on Water Pollution due to Toxic Metals in Ulhas River Flowing along the Dombivli City of Mumbai India. *Int Lett Nat Sci*. 2015;38:66–76. doi:10.18052/www.scipress.com/ILNS.38.66.
12. Shunmugam AP, Subramanian G, Fernandez JG. Measurements of the swimming speeds of motile microorganisms using object tracking and their correlation with water pollution and rheology levels. *Sci Rep*. 2021;11:11821. doi:10.1038/s41598-021-91134-1.

Author biography

Hiba Mohd Shujaat Shaikh, EA to Director

Mukesh Pimpliskar, HOD https://orcid.org/0000-0002-4482-228X

Cite this article: Shaikh HMS, Pimpliskar M. Effect of heavy metals on *paramecium* found in different water bodies of Bhiwandi. *IP Int J Med Microbiol Trop Dis* 2022;8(2):127-131.