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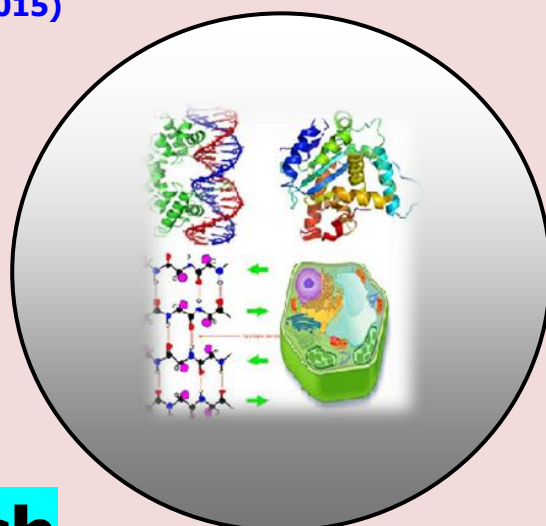
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Atomic Absorption Spectroscopy Detection of Heavy Metals (Pb, Hg, As, Cu) Contamination in the Water of Natural Lakes of District Gorakhpur, U. P. India**Pooja Agrahari¹, Vishnu Pragya Parmita Singh², Archana Singh², Archana Singh², V. K. Singh¹ and D. K. Singh*¹**¹Malacology Laboratory, Department of Zoology, D.D.U. Gorakhpur University, Gorakhpur, U.P. 273009, India.²Department of Environmental Science, D.D.U. Gorakhpur University, Gorakhpur, U.P. 273009, India.**ABSTRACT**

Heavy metal contamination in natural water reservoir has been recognized as a major environmental concern due to their pervasiveness and persistence. These heavy metals attain higher concentrations due to bioaccumulation in different part of plants and finally pose serious hazard to animal/human. These heavy metals are non- biodegradable especially Cu, Hg, Pb; hence there is a need to develop such remediation technique, which would be efficient, economic and rapidly deployable in a wide range of physical settings. Detection of heavy metals in water of natural Lake Ramgarh had large content of Mercury (0.003 mg/L), Lead (0.03 mg/L), and Copper (0.139 mg/L). In other natural lake Maheshara, the amount of Hg, Pb and Cu in water was 0.0003 mg/L, 0.05 mg/L and 0.001 mg/L, respectively whereas in Suraj Kund lake, the amount of Hg, Pb and Cu in water was 0.0009 mg/L, 0.05 mg/L and 0.001 mg/L, respectively. The soil contaminated by these effluents containing water and consumption of fishes of these natural lakes could be hazardous to human beings and their health.

Keywords: Arsenic; Atomic Absorption Spectroscopy; Copper; Heavy metals; Lead; Mercury.

INTRODUCTION

Heavy metals, in contrast to organic materials, cannot be degraded, and accumulate in water, bottom sediments and living organisms (Miretzky *et al.*, 2004). Water contamination with heavy metals is a very important problem in the current world. WHO (2002) estimates that about a quarter of the human disease today occur due to prolonged exposure of environmental pollution (Azimi and Sadeghi, 2013). Occurrence of toxic metals in lake, pond, ditch and river water affect the lives of local people that depend upon these water sources (Rai *et al.*, 2002). Consumption of such

aquatic foodstuff enriched with toxic metals may cause serious health hazards through food chain magnification (Khan *et al.*, 2000). Water pollution (groundwater, lakes, streams and rivers) occurs due to three major reasons i.e. drainage of excess nutrients from sewage, wastes from industries and mining and agriculture (Jamode *et al.*, 2004). Acid rain can exacerbate this process by releasing heavy metals trapped in soils. Plants are exposed to heavy metals through the uptake of water; animals eat these plants; ingestion of plant- and animal-based foods is the largest sources of heavy metals in humans (Radojevic and Bashkin, 1999). Some of the metals are necessary to present in drinking water up to desirable limit. However, their higher concentrations i.e. more than permissible limit are toxic for human beings (WHO, 2011; BIS, 2012; CPCB, 2013). In the present study, we selected the three famous ponds and lakes of Gorakhpur city, because effluents that are continuously released by industries, factories, domestic waste etc. have polluted these water bodies, disturbing the quality of water also degrade the protein source in the form of fish food and limit their use. In future, we have also studied the bioaccumulation of these metals in fish, which are live in that habitat.

The aim of the present study is the assessment of heavy metals As, Pb, Hg, and Cu by atomic absorption spectroscopy in the water of different lakes of Gorakhpur.

MATERIALS AND METHODS

Study Area

Water of natural lake i.e. Ramgarh, Maheshara and Suraj Kund Pond were collected to study the level of heavy metals. All these lakes/ponds were situated in Gorakhpur (Latitude 26° 46' N, Longitude 83° 22' E) district of Uttar Pradesh, India on the left bank of river Rapti (Figure 1). Gorakhpur is an industrialized city, heavy a population of many lakh. Different types of industries effluent along with municipal waste are dumped in these Lakes of Gorakhpur city.

Ramgarh Lake is a lake covers an area of 723 hectares (1,790 acres) with a circumference of 18 kilometers on the southeast of Gorakhpur city. The lake supports a significant fishery on which numerous people depend upon it for their livelihood. The Lake water is also used for irrigation and recreation. However, the lake has deteriorated rapidly because it receives huge amount of domestic wastewater and solid wastes from the urban catchment. Large-scale fish kills have been reported frequently in recent years. The lake also supports a large diversity of macrophytes but their excessive growth, and particularly that of water hyacinth, has created serious problems of water quality as well as for its use.

Likewise, Ramgarh Lake, **Maheshara Lake** is also an important water body of Gorakhpur District. It is a tributary of Rapti Lake. Maheshara Lake is a natural habitat Lake and slightly attached with River Rapti of Gorakhpur District. Availability and quality of fresh water in this lake is important because it provide employment to local angler and main source of livelihood of some of very poor community of this area. At present, Maheshara Lake is polluted due to discharge of effluents from industries, domestic sewage and municipal waste bodies washing from agricultural land using pesticides and chemical fertilizers (Srivastava and Singh, 2014). Income from capture fishery is main source of livelihood of some of very poor community. It is obvious that major food fishes are under threat and needs urgent strategic conservation effort. Lack of information in this field has become an obstacle in the development of long run conservation and recovering process (Jain, 2009).

The **Suraj Kund** is an ancient man made Pond. It is about 20 feet deep and is surrounded by Ghats. It is a focal point of various types of religious and cultural practices throughout the year. Thousands of people gather near the pond and offer their worship by virtue of religious bathing and offering worship materials in the form of flowers, garlands, banana plant, painted earthen pots, oil, earthen lamps etc.

Notably, during Deepdan held the next day of Deepawali festival, about 25,000 earthen lamps are lighted around the pond. The spilling of vegetable oil could eventually, lead to its spreading on the water surface, thereby, preventing the exchange of gases with atmosphere, while also causing organic pollution in the pond. Fish kills have also been reported in the pond on some occasions.

Sampling procedure

The water samples from Ramgarh, Maheshara Lake and Suraj Kund Pond were done belong 10 feet in depth from the water surface. Water samples were collected from the six sampling points of each pond and lakes and stored in clean water sampler bottles. These samples were acidified immediately with 2 ml of HNO₃ per liter of water and then transported to the laboratory and preserved at 4°C until subsequent analysis. These samples are filtered through a Whatman glass microfiber filter (GF/C) to avoid any disruption in the instrument.

Standards Used

Standards of Hg, Pb, As and Cu were purchased from Perkin Elmer, 710 Bridgeport Avenue (USA).

Analytical Methods

A flow injection-mercury hydride system technique was used for the quantitation of As and Hg, and a graphite furnace technique was used for the analysis of Pb and Cu. A Perkin Elmer Atomic absorption spectrometer AAnalyst 400 was used, coupled with graphite furnace controller. The two heavy metals (Hg, As) standard reagents preparation is done by Mercury hydride System (MHS 15) user guide of Perkin Elmer. The MHS-15 Mercury Hydride System is a manual accessory for high-sensitivity determination of mercury and hydride-forming elements, such as As, Se, Sb, Te, Bi and Sn, by flame Atomic Absorption Spectrometry (AAS). The analytical wavelengths were at 283.31 for Pb, 324.75 for Cu, 253.65 for Hg, and 193.70 for As. At least five standard solutions were prepared for each metal detection.

Preparation of chemicals

Mercury content in water was followed by cold vapour atomic absorption spectrometry method (APHA, 2005). Standard reagents for mercury were prepared with 1.5% w/v HCl, 1.5% v/v HNO₃, 5% (w/v) KMnO₄ solution, 1% (w/v) NaOH solution and 3% (w/v) NaBH₄ solution. NaBH₄ or sodium tetrahydroborate is used as a reductant. The stock solution contains 1000 mg/L Hg. 1 ppm standard Hg was prepared by using 100 ml from 1000 ppm dilute in 1.5% HCl. Small amount of 1.5% HCl was taken and add calculated amount of standard from 1000 ppm, and shake well. Then add 3-4 drops of 5% KMnO₄ solution and then make up to 100 ml from 1.5% HCl. Then 1 ppm Hg standard was used as stock and 10 ppb, 20 ppb, 25 ppb and 50 ppb standard were prepared by this stock solution in 1.5% HCl.

Standard reagents preparations for arsenic were prepared with 1.5% (v/v) HCl, 1% (w/v) NaOH Solution, **Reductant solution:** 3% (w/v) NaBH₄ Solution (making in 1% NaOH solution), **Reducing Agent:** KI solution (KI + Ascorbic Acid). Standard Solution of arsenic was prepared by taking 10 ml 1.5% v/v HCl solution in 3 volumetric flasks. Standard of 2, 4 and 8 ppb was taken, and then 10 ml Ascorbic acid- KI solution was added, and then it was kept for 45 minutes for pre-reduction. After 45 minutes, it was made to 100 ml.

Atomic absorption spectrometer (AAS) was calibrated by running the standard solution and calibration curves were obtained. Thereafter the unknown solution was fed into the instrument and the absorbance of the element present in the solution was measured. The concentration of heavy metals in unknown solution was calculated from the calibration curve. In case of Copper and Lead, only standard solutions were used for the analysis of the samples.

RESULTS

The samples collected from three sites (Figure 1) were tested for the detection of the metals Copper, Arsenic, Mercury and Lead. All readings are the mean value of metal level. In Ramgarh Lake water sample, level of copper is 0.139 ± 0.03 mg/L, whereas concentration of copper in Maheshara Lake and Suraj Kund pond was 0.001 ± 0.004 mg/L (Figure 2).

Mercury concentration noted in Ramgarh Lake was 0.003 ± 0.017 mg/L. comparatively; the concentration of mercury in Maheshara Lake and Suraj Kund pond was 0.0003 ± 0.002 mg/L and 0.0009 ± 0.00 mg/L, respectively (Figure 3).

Concentration of Arsenic in Ramgarh Lake was 0.002 ± 0.005 mg/L, while in Maheshara Lake and Suraj Kund pond was 0.001 ± 0.002 mg/L (Figure 4).

Concentration of Lead in Ramgarh Lake was 0.03 ± 0.01 mg/L, while in Maheshara Lake and Suraj Kund pond was 0.05 ± 0.04 and 0.05 ± 0.017 mg/L, respectively (Figure 5).

DISCUSSION

India today is one of the first ten industrialized country of the world. Today we have good industrial infrastructure. Consequently, vast amount of pollutants contaminate the environment. With the increasing use of a wide variety of metals in industry and in our daily life, it assumed serious environmental hazards of different dimensions were noticed (Singh, 2005). The impact of these heavy metals on human health is currently an area of intense interest due to the ubiquity of exposure. Many studies have so far been published on heavy metal contamination because of their effects on human health and ecosystem (Caussy *et al.*, 2003; Buschmann *et al.*, 2008; Mousavi *et al.*, 2013). Metal containing industrial effluents constitute major source of metallic pollution of Hydrosphere. Pb, Zn, Cu, Mn, Co, Ni, Cd, Cr, and Mo are toxic and carcinogenic agents reported in drinking water of many areas around the world (Groopman *et al.*, 1985).

Heavy metals are of concern due to their biodegradable nature and persistence in the natural environment. They have the ability to bio-accumulate in aquatic ecosystems (Miller *et al.*, 2002) and are capable of transforming into more toxic compounds. Metals are well known for their property of accumulation in the human body and their synergistic effects to organisms. Some metals are also known for their teratogenic and carcinogenic effects (Csuros and Csuros, 2002). Fishes have great propensity for bio-accumulation and magnification of heavy metals (Obasohan *et al.*, 2006), thus fish contamination in the aquatic environment by heavy metals is a potential threat to human health. Fishes become enriched with accumulated heavy metals because aquatic microflora and microfauna, which constitute fish foods, are capable of accumulating heavy metals in their living cells (Obasohan *et al.*, 2006).

The level of copper in Ramgarh Lake water was 0.139 mg/L that was almost double the permissible limit of ISI (0.05 mg/L) but less than the WHO limit, which are 1.0 mg/L. It indicates that the water body is heavily polluted with the copper metal. The various sources of metal contaminations are the industrial effluents, metallic mining, and metallurgical processes, vehicle washes from nearby shops, domestic garbage's and other waste materials dumped by the surrounding inhabitants. If the level of copper ion is exceeded, then the immediate health effects vomiting, diarrhoea, stomach cramps and nausea will be possible. The effects are much higher in children under one year old than adults. However, long-term exposure, which is more than 14 days to copper in the drinking water, can cause serious problems like kidney and liver damages in infants (Hutzinger, 1995; Soylak *et al.*, 1998).

The British physician **Dr. Samuel Alexander Kinnier Wilson** (1878-1937), a neurologist described the copper toxicity condition. Since then this disease is called **Wilson's disease**. Wilson's disease or hepato lenticular degeneration is associated with copper accumulation in tissues. This manifests as neurological, psychiatric, or liver disease and occasionally liver transplant is required.

The intake of copper salts in larger quantities may cause haemolysis, hepatotoxic, neurotoxic effects, gastrointestinal, hepatic, and renal effects with symptoms such as severe abdominal pain, vomiting, diarrhoea, hepatic necrosis, hematuria, proteinuria, hypotension, tachycardia, convulsions, coma, and death (Tchounwou *et al.*, 2008). The high level of copper in Ramgarh Lake indicates that the water body is severely contaminated with copper and serious action should be taken within no time so as to relieve the water body from contamination.



Figure 1 Schematic representation of various sampling sites from Gorakhpur city. The symbols refer to the following sampling sites:

- ↓ Mahesara Lake
- ★ Suraj Kund
- Raamgarh Taal

Level of mercury in Ramgarh Lake is 0.003 mg/L, which is almost thrice the permissible limit of WHO (0.001 mg/L). This indicates that Ramgarh Lake is highly polluted with mercury. This condition is alarming which can result bioaccumulation of Hg in fishes inhabiting the Lake. However, the level of mercury in Maheshara Lake (0.0003 mg/L) was very low. This indicates that there is no any sign of contamination in them. Nevertheless, it should be checked regularly to avoid any contamination. In Suraj Kund pond (0.0009 mg/L) the level of mercury are at par with the permissible limit of WHO, ISI and ICMR which is 0.001 mg/L. All mercury species is toxic, with organic mercury compounds generally being more toxic than inorganic species (Leopolda, 2010). Organomercuric compounds reduce the photosynthetic ability of phyto-planktons and zooplanktons of freshwater and marine ecosystems. However, inorganic mercury cannot cross the blood- brain barrier but reaches the kidney and cause damage to it. Hg_2^{2+} forms an insoluble chloride with chloride ion. Though it is not toxic but because of its high affinity for sulphur atoms, it easily attaches to the sulphur containing amino-acids of proteins.

It also forms bonds with haemoglobin and serum albumin, both of which contain sulphhydryl groups. However, this ion does not travel across the biological membranes.

As it is well known that, the mercury has the tendency to accumulate in the food chain and it is magnified as it passes to upper trophic levels. Acute oral mercury poisoning results primarily in haemorrhagic gastritis and colitis; the ultimate damage is to the kidney (WHO, 2005). The mercury level in the Maheshara Lake (0.0003 mg/L) is very much below the permissible limit but proper monitoring is needed in order to avoid any further contamination in future. However, the level of mercury in Ramgah Lake (0.003 mg/L) and Suraj Kund Pond (0.0009 mg/L), which has already reached to the water sources, will pose its harmful effects for a long time.

Level of arsenic is much below in comparison to maximum permissible limit in all the water bodies. However, the As level in Ramgarh is slightly higher than the other two water bodies. Arsenic level is not in alarming situation in water of these lakes, but it should be regularly monitored. The level of lead in the Ramgarh is (0.03 mg/L) against the maximum permissible limit which is 0.05, however in case of Maheshara and Suraj Kund it is about 0.05mg/L is just at par with the permissible limit. The state is alarming as the lead contamination is just on the verge and if measures are not taken to control it, the condition will be worsened. Exposure to lead is very dangerous for young children compared to an adult. This is because young children's growing rate is much higher than adult. Lead can accumulate in human body over some time and cause serious damage to brain, kidney, nervous and red blood cells. For infants, large amount of lead can cause delays in physical and mental development (Hutzinger, 1995). The main sources of lead contamination in the environment are the industrial discharges from smelters, battery-manufacturing units, run-off from contaminated land areas and sewage effluents.

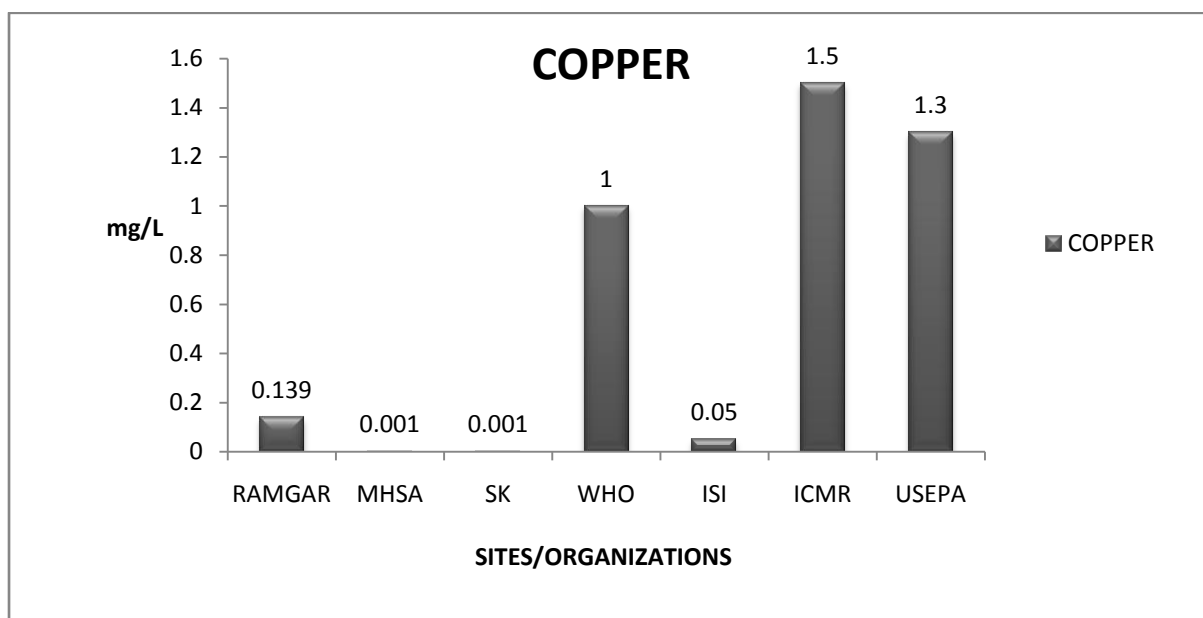


Figure 2 Level of copper found in different water samples and maximum permissible limits of various health organizations.

Lead is known to disrupt several enzymes. The major biochemical effect of Pb is its interference with heme- synthesis, which leads to hematological damage (Reddy, 2005). Pb inhibits several enzymes involved in heme synthesis. Lead interferes at several steps in heme (the pigment that combines with protein to form haemoglobin)

synthesis in the bone marrow by inhibiting the activities of enzymes ferrochelatase, α - amino laevulinate dehydrogenase (ALAD) and with the uptake of iron into mitochondria (Piomilli, 1980). It is a cumulative general poison and associated with several health hazards like anaemia (Moore, 1988) which leads to deficiency of haemoglobin and reproductive effects (Wildt *et al.*, 1983; Cullen *et al.*, 1984).

ABBREVIATIONS- RMGH= Ramgarh Lake, MHSA= Maheshara Lake, SK=Suraj Kund, WHO= World Health Organization, ISI= Indian Standard Institute, ICMR= Indian Council of Medical Research, USEPA= United States of Environmental Protection Agency, mg/L= milli grams per liter.

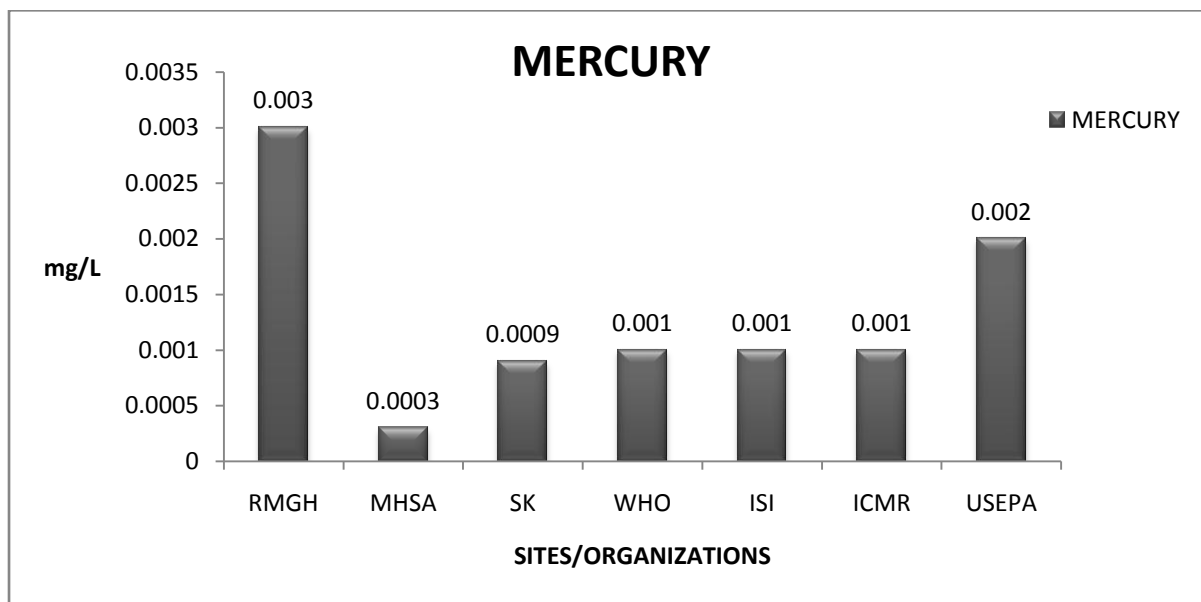


Figure 3 Levels of mercury in different water samples and maximum permissible limit of various health organizations.

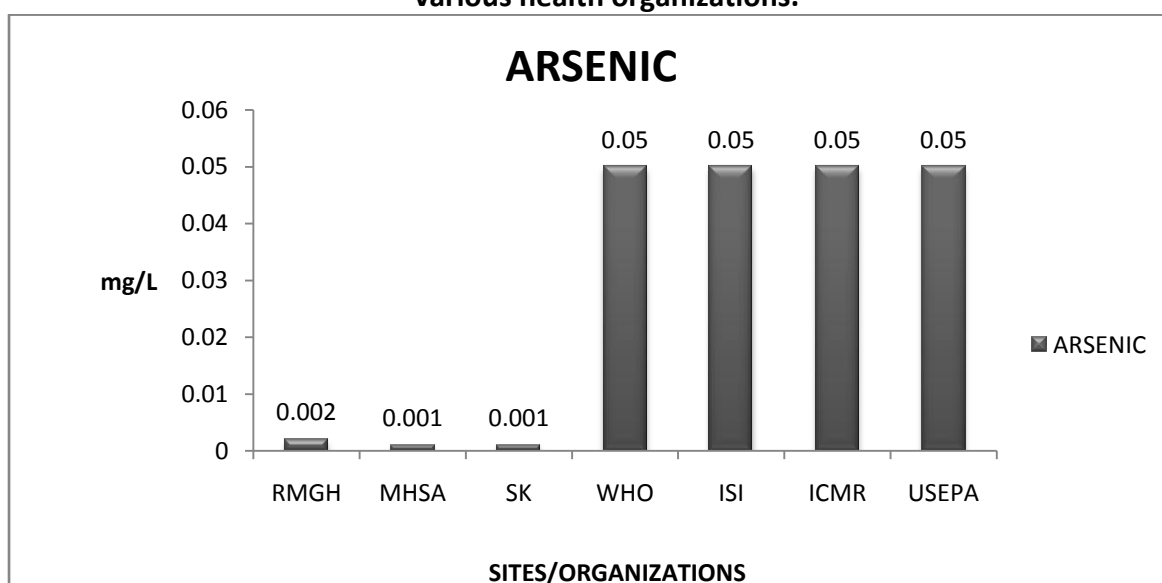


Figure 4 Level of Arsenic in different water samples and maximum permissible limit of various health organizations.

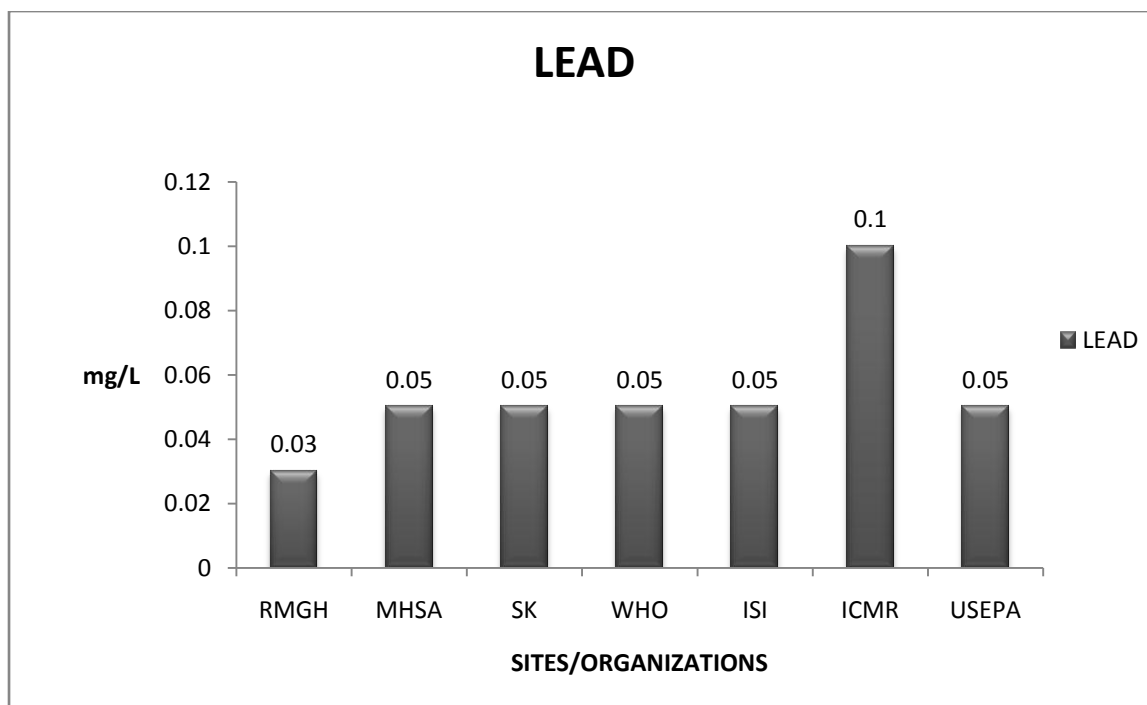


Figure 5 Level of Lead in different water samples and maximum permissible limit of various health organizations.

CONCLUSIONS

The level of different metal concentrations viz. Copper, Mercury, Arsenic, Lead obtained in different water samples clearly indicate that these water bodies are contaminated with metals. There is a need for quick action to be taken so that further contamination could be prevented. Uncontrolled disposal of solid wastes and incineration of solid waste should be banned and responsible authorities have to monitor and control the contamination to these water bodies periodically. The solid waste should be managed properly either by landfills, composting or incineration. Their dumping into the water body should be strictly avoided. However, this can only be possible with proper awareness plans which could be conducted among the local inhabitants. The cleaning programme should be undertaken and further dumping should be avoided. Concentration of soil heavy metals above the threshold level indicate that their measurement in fishes inhabiting these lakes as well as plants and vegetables irrigated with water of these lakes should be measured to note the current status of metal pollution.

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