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Statistical analysis of hydrological properties and genetic toxicity of Maheshara Lake

Pallavi Srivastava and Ajay Singh*

Department of Zoology, D.D.U.University Gorakhpur, India.

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The purpose of this study was to assess the water qualities and genotoxicity of Maheshara Lake situated near Gorakhpur, India. The lake receives many types of pollutants from two industries, agricultural run-off and domestic sewages. A total of five sampling stations were established for measurement of water pollution and the measurement of cytogenetic alterations *in vivo* is considered an initial step in the risk assessment procedures for genotoxic agent. A study was conducted to assess the cytogenetic changes in an airbreathing fish, *Channa punctatus* inhabiting the polluted water of Maheshara Lake. In the present study, significantly ($p<0.05$) higher micronuclei formation was observed in these groups of fishes which receive lake water thus indicating induced mutation in fishes living in Maheshara Lake water. It might be the presence of genotoxic chemicals in water, which is responsible for the DNA damages in fishes.

Key words: Maheshara Lake, genotoxic, micronuclei, *Channa punctatus*.

INTRODUCTION

In nature, water is one of the most abundantly available resources, which has been exploited by the man. Water is considered as a good disposable site of city sewage and also of industrial wastage. Quality of water provides current information about the concentration of various solutes at a given place and time. In this fast growing age, declining availability of usable fresh water is the major concern in terms of water quantity and quality (Boyd and Tucker, 1998)

In toxicity test, the genotoxicity test in fishes, amphibians and other aquatic organisms serves as an effective tool in monitoring environmental quality. In aquatic ecosystem, genotoxic pollutions refer to the introduction of contaminants having mutagenic, teratogenic and carcinogenic potentials into its principle media (Fagr et al., 2008). The compounds like heavy metals (Matsumoto et al., 2005), microbial toxins (Smith, 1996) and pesticides as fungicides (Srivastava and Singh, 2013), insecticides and so on are considered as genotoxicants that come from indiscriminate disposal of sewage, industrial wastes and human activities. These genotoxicants have been reported to cause mutations because of their strong affinity to bond with DNA and resulting in the formation of DNA adducts and micronuclei. These genomic losses in organisms induce various disease. Therefore, now a day detection of genotoxic effects in aquatic organisms has gained importance (Hayashi et al., 1998; Kushwaha et al., 2012).

The micronucleus (MN) test, due to its simplicity, is one of the most applicable techniques to identify genomic alterations in environmental animals (Hayashi and Bolognesi, 2011). Micronuclei express in dividing cell that either contains chromosome breaks lacking centromere or whole chromosomes, which unable traveling to spindle poles during mitosis. At telophase, a nuclear envelope forms around the lagging chromosome and fragments which uncoil and gradually assume morphology of an interphase nucleus. With an exception, they are smaller than the main nuclei in the cell, hence termed as micronuclei.

*Corresponding author. E-mail: 5september1984@gmail.com. Tel: 91-5512202127.

Fishes response to toxic agents similar to higher vertebrates can allow the assessment of substances that are potentially hazardous to humans (Grisolia and Carderio, 2002; Kushwaha et al., 2012). Due to chemical pollutants, the low amount of DNA per cell, the large numbers of small chromosomes, and the low mitotic activity in many fish species impaired the metaphase analysis of chromosomal damage and sister chromatid exchanges are demonstrated (Pavlica et al., 2000). The MN test, due to its potentiality to be applied in any proliferating cell population regardless of the karyotype of the species use, are successfully applied in fish for evaluating the genotoxic activity of xenobiotic agents and of complex environmental mixtures in laboratory as well as in field studies (Hayashi and Bolognesi, 2011).

The Gorakhpur City lies between Lat. 26° 13´N and 27° 29´N and Long. 83° 05´ E nad 83° 56´E near the bank of River Rapti. The Maheshara is a tributary of Rapti, is an important water body of Gorakhpur, India region. In this region, people use synthetic pesticides for pest control in agricultural fields especially in paddy fields, which received the effluents of the two industries (metal workshop and fertilizer enterprises), domestic sewage and agricultural run-off. Lake water is also used for bathing, cleaning of utensils, washing of cloths and disposable of industrial wastage. These activities have been subjected to a strong biotic pressure all along the periphery and have turned into an eutrophic stage. The water of the lake is declining in quality posing threat to these people. It has also noticed that the highly polluted water of Maheshara Lake affect the population of aquatic fauna especially fishes. Therefore, the study on the fish genotoxicity in Maheshara Lake was lacking, hence the present study had aimed at examining the water quality and fish genotoxicity of lake river system in Gorakhpur, India. The findings from the study will benefit the planning and management of sustainable fisheries and conservation of natural resources.

MATERIALS AND METHODS

Site selection

Five study sites were selected as shown in Table 1. Site 1 is agricultural runoff point. This site was chosen because there was paddy fields surrounding the lake and local farmers had used several pesticides in the fields and by surface run off these pesticides enter into body tissues of fishes and alter the biochemical pathways and also induce genetic disorders, Site 2 was metal workshop point selected because the effluents of this industry continuously reach in the lake, Site 3 was domestic water point, lake water has also been used for keeping house hold activities by the surrounding people who are inhabitant, Site 4 was sewage point, most of the city sewage discharge into the lake and site 5 was industrial effluent point. There was another industry, which effluents also reach in the lake.

Collection of water sample

In Gorakhpur city, seasonal variations were found. May-June have extremely hot day while November-December are extremely cold day and July-August is a rainy month. Hence, water samples were collected in these months. Water samples were collected early in morning (6:00to 8:00am) from May-June (2013), July-August (2013) and November-December (2013). From each site, 10 samples of water were collected and averages of each site are presented in Table 1. Temperature and pH of water samples were measured in the field immediately with the help of mercury glass thermometer and portable pH meter, respectively. Other physico-chemical parameters were analyzed in the laboratory within 5 h of collection. All the parameters such as dissolved oxygen (DO), biochemical oxygen demand (BOD), chemical oxygen demand (COD), total solids and total dissolved solids were examined by using standard methods APHA (2005).

Collection of fishes

For genotoxic experiment, fishes were collected with the help of expertise of local fisher folk. Sampling was carried out at all the five sites. Different types of gear including cast net, gill net and drag net and other local conveners were used. *Channa punctatus* is edible fish and mostly found in lake at all sites.

Experimental designs for micronucleus test assay (MNT)

C. punctatus were collected from different sites of Maheshara Lake. For experiment, 9 fishes were collected from all sites and divided into three groups. An average number of micronuclei is shown in Table 2.

Slide preparation

Blood smear slides were prepare by the method of Das and Nanda (1986) with some modifications. Peripheral blood samples were obtained from caudal vein of fish. For each groups, fishes used and blood smear were prepared. At each assessment, 2500 cell/fish were analyzed, 7500 erythrocytes for each group. Slides were dried in air, fixed in absolute methanol for 10-15 min, and stained in Giemsa (pH 7.0) for 1-2 h; washed with DDW and air-dried. Permanent preparation was made by mounting in DPX. They were screen in oil immersion objective (100x) and micronuclei was observed under microscope (OLYMPUS CX21i) with camera (Magnus MIPS USB 5MP).

Chemicals and Instruments

Chemicals such as Di-sodium hydrogen orthophosphate (17549) from Merck chemicals, Potassium Di-hydrogen orthophosphate (39619K05) from Merck chemicals, Giemsa stain (44034G25 G09Y/1009/0907/71) from SD Fine chemicals India, Glycerol, Xylene and Methanol from SD Fine chemicals India were used. The instruments used in the study are Microscope (OLYMPUS CX21i) purchased from Japan, Camera used in the experiment (Magnus MIPS USB 5MP) was purchased from Japan, Hot Air Oven (LIMCO equipment Limited).

Statistical analysis

The frequencies of abnormalities were determined for each group. MN frequencies (MN‰) are calculated as follows:

$$MN‰ = \frac{\text{Number of cells containing micronucleus} \times 1000}{\text{Total number of cells counted}}$$

Table 1. Physico-chemical parameters of water sample at different sites of Maheshara Lake.

Parameter	May-June					July-August					November-December				
	Site 1	Site 2	Site 3	Site 4	Site 5	Site 1	Site 2	Site 3	Site 4	Site 5	Site 1	Site 2	Site 3	Site 4	Site 5
pH	8.02±0.11	6.25±0.05	10.14±0.12	10.04±0.10	10.0±0.09	7.92±0.10	5.15±0.01	7.80±0.07	7.50±0.07	7.85±0.06	7.10±0.06	5.57±0.02	7.0±0.05	6.91±0.04	7.15±0.06
Temp	32.2±0.10	35.3±0.12	37.0±0.20	35.41±0.18	35.0±0.16	30.0±0.09	32.4±0.11	34.1±0.12	32.0±0.10	31.5±0.13	26.12±0.10	28.5±0.14	28.0±0.13	28.71±0.12	28.0±0.10
DO	2.85±0.13	1.80±0.09	1.95±0.10	2.0±0.12	1.82±0.09	5.20±0.12	4.61±0.09	4.90±0.10	5.10±0.26	4.0±0.06	6.0±0.17	5.60±0.14	5.85±0.15	6.03±0.18	5.35±0.12
BOD	20.33±0.14	40.0±0.20	30.40±0.18	41.66±0.24	25.0±0.16	28.35±0.17	33.3±0.20	31.03±0.19	35.56±0.22	30.54±0.18	31.78±0.19	36.29±0.23	33.04±0.20	39.83±0.28	32.29±0.21
COD	82.10±1.02	120.23±1.14	100.43±1.01	125.56±1.12	90.25±1.14	113.34±1.16	133.20±1.09	124.12±1.14	142.27±1.05	122.19±1.12	127.14±1.10	145.17±1.06	132.19±1.04	159.32 ±1.07	129.16±1.13
TSS	317±1.11	303±1.13	447±1.24	353±1.15	385±1.20	213±1.08	235±1.13	278±1.10	245±1.12	274±1.14	184±1.06	198±1.09	210±1.13	201±1.10	206±1.11
TDS	154.25±1.17	150.0±1.12	186.75±1.20	163.25±1.15	171.25±1.17	193.12±1.19	187.32±1.20	232.32±1.34	200.23±1.28	210.56±1.22	220.15±1.25	212.32±1.21	265.34±1.42	228.45±1.30	236.12±1.32

DO = Dissolve oxygen (mg/l), BOD = biological oxygen demand (mg/l), COD = chemical oxygen demand (mg/l), TSS = total suspended solids (mg/l), TDS = total dissolve solids (mg/l), Temp=Temperature (°C).

Then the mean ± standard error for each group was calculated. Student's t-test was employed for compareson of control and experimental animals.

Correlation coefficients between different pairs of parameters were calculated by using the following formula and t-test was applied for checking significance.

$$r = \frac{n(\Sigma xy)-(\Sigma x)-(\Sigma y)}{\sqrt{[n\Sigma x^2-(\Sigma x)^2][n\Sigma y^2-(\Sigma y^2)]}}$$

Where n = sample size.

RESULTS

Physico-chemical analysis

The physico-chemical characteristics of Maheshara Lake during the period of study at different sites are presented in Table 1.

pH

In the present study during May-June, the highest pH (10.14 ± 0.12) was recorded at Site-3 (domestic wastewater point) while lowest pH (6.25 ± 0.05) was recorded at Site-2 (metal workshop point). During July-Aug the highest pH (7.92 ± 0.10) was recorded at Site-1 (agriculture run off point) while lowest pH (5.15 ± 0.01) was recorded at Site-2 (metal workshop point). During November-December the highest pH (7.10 ± 0.06) was recorded at Site-1 (agriculture run off point) while lowest pH (5.57 ± 0.02) was recorded at Site-2 (metal workshop point).

Water temperature

Water temperature was a controlling factor for aquatic life. During May-June the highest temperature (37.0 ± 0.20°C) was recorded at Site-3 (domestic wastewater point) while lowest temperature (32.2 ± 0.10°C) was recorded at Site-1 (agriculture run off point). During July-Aug the highest temperature (34.1 ± 0.12°C) was recorded at Site-3 (domestic wastewater point) while lowest temperature (30.0 ± 0.09°C) was recorded at Site-1 (agriculture run off point). During November-December, the highest temperature (28.71 ± 0.12°C) was recorded at Site-4 (sewage waste point) while lowest temperature (26.12 ± 0.10°C) was recorded at Site-1 (agriculture run off point).

Dissolve oxygen (DO)

Dissolve oxygen was one of the most important parameters. Concentrations of dissolved oxygen support a great diversity of aquatic organisms. During May-June the highest concentration of DO (2.85 ± 0.13mg/l) was recorded at Site-1 (agriculture run off point) while lowest concentration of DO (1.80 ± 0.09 mg/l) was recorded at Site-2 (metal workshop point). During July-August, the highest concentration of DO (5.20 ± 0.12 mg/l) was recorded at Site-1 (agriculture run off point) while lowest concentration of DO (4.0 ± 0.06 mg/l) was recorded at Site-5 (industrial provision point). During November-December, the highest concentration of DO (6.03 ± 0.18 mg/l) was recorded at Site-4 (sewage waste point) while lowest concentration of DO (5.35 ± 0.12 mg/l) was recorded at Site-5 (industrial provision point).

Biological oxygen demand (BOD)

Biological oxygen demand test show how much oxygen is being consumed. During May-June the highest value of BOD (41.66 ± 0.24 mg/l) was recorded at Site-4 (sewage waste point) while lowest value of BOD (20.33 ± 0.14 mg/l) was recorded at Site-1 (agriculture run off point). During July-August the highest value of BOD (35.56 ± 0.22 mg/l) was recorded at Site-4

Table 2. Correlation between different pairs of parameters.

Pair of parameter	r-value	t-value
pH and temp.	0.4020	7.60
pH and DO	-0.0112	1.95
pH and BOD	0.0884	1.53
pH and COD	-0.1516	2.65
pH and TSS	0.8320	25.9
pH and TDS	0.8434	26.9
DO and BOD	-0.3459	6.38
DO and COD	-0.5854	12.5
DO and TSS	-0.3428	6.32
DO and TDS	-0.3063	5.57
BOD and COD	0.9984	31.27
BOD and TSS	-0.1557	2.73
BOD and TDS	-0.1673	2.93
COD and TSS	-0.1815	3.19
COD and TDS	-0.1905	3.36
TSS and TDS	0.9998	86.58

(sewage waste point) while lowest value of BOD (28.35 ± 0.17 mg/l) was recorded at Site-1 (agriculture run off point). During November-December, the highest value of BOD (39.83 ± 0.28 mg/l) was recorded at Site-4 (sewage waste point) while lowest value of BOD (31.78 ± 0.19 mg/l) was recorded at Site-1 (agriculture run off point).

Chemical oxygen demand (COD)

On other hand, Chemical oxygen demand (COD) range during May-June was highest (125.56 ± 1.12 mg/l) at Site-4 (sewage waste point) lowest (82.10 ± 1.02 mg/l) Site-1 (agriculture run off point). During July-August, the highest value reached up to (142.27 ± 1.05 mg/l) at Site-4 (sewage waste point) lowest (113.34 ± 1.16 mg/l) at Site-1 (agriculture run off point). During November-December, the highest reached up to 159.32 ± 1.07 mg/l at Site-4 (sewage waste point) while lowest value reached up to 127.14 ± 1.10 mg/l at Site-1 (agriculture run off point).

Total suspended solids

During May-June, Total suspended solids recorded from (303 ± 1.13 mg/l) to (447 ± 1.24 mg/l). During July-August, it recorded as from 213±1.08 mg/l to 278 ± 1.10 mg/l. During November-December, it recorded from 184 ± 1.06 to 210 ± 1.13 mg/l. Total dissolve solids was from 150.0±1.12 to 186.75 ± 1.20 mg/l during May-June. During July-August, it was from 187.32 ± 1.20 to 232.32 ± 1.34 mg/l. During November-December, it was from 212.32 ± 1.21 to 265.34 ± 1.42 mg/l.

Micronuclei assay

The result of micronucleus in peripheral erythrocytes of *C. punctatus* at different sites of Maheshara Lake are shown in Table 3 and Figure 1. At Site 1, which was agricultural runoff point, the micronuclei frequencies was recorded as 9.60 ± 0.04. At Site 2, The metal workshop point, induction of micronuclei in fish *C. punctatus* was observed as 13.33 ± 0.05. Site 3, domestic water point, the induction of frequencies in peripheral blood samples were 19.86 ± 0.08. Site 4 was sewage point and the induction of frequencies in peripheral blood samples were also 22.13 ± 0.09, and At Site 5, the Industrial effluent point observed frequency was 13.86 ± 0.06. In all the groups receiving polluted water, frequencies of micronuclei were observed to be significantly higher ($p<0.05$) when compared with the control. The maximum induction in micronuclei frequency was observed at Site 4 and minimum was observed at Site 1. The hierarchy of micronuclei frequencies at different sites were as follows: Site 4 > Site 3 > Site 5 > Site 2 > Site 1 as compared to the control.

DISCUSSION

Different water quality parameters with significant correlation coefficients are given in Table 2.

pH

pH is an important limiting chemical factor for aquatic life. The H^+ or OH^- ion activity may disrupt aquatic organisms biochemical reactions by either harming or killing the lake organisms. Correlation analysis showed that pH had a significant positive relationship with temperature (r = 0.4020, t = 7.60), BOD (r = 0.0884, t = 1.53), TSS (r = 0.8320, t = 25.9) and TDS (r = 0.8434, t = 26.9) and negative correlation between D.O. (r = 0.0112, t = 1.95) and

Table 3. Frequencies of micronuclei (MN) observed in fish *C. punctatus* at different sites of Maheshara Lake.

Site	Total number of cells studies	Total number of MN			Average percentage (%) aberration* ±SE
		Group 1	Group 2	Group 3	
Control (freshwater fish)		02	01	01	0.8±0.05
Site 1		21	28	23	9.60±0.04
Site 2	2500cell/fish	34	30	36	13.33±0.05
Site 3		50	46	53	19.86±0.08
Site 4		56	53	57	22.13±0.09
Site 5		38	32	34	13.86±0.06

*, Significant (P<0.05) when Student's 't' test was applied between treated and control groups.

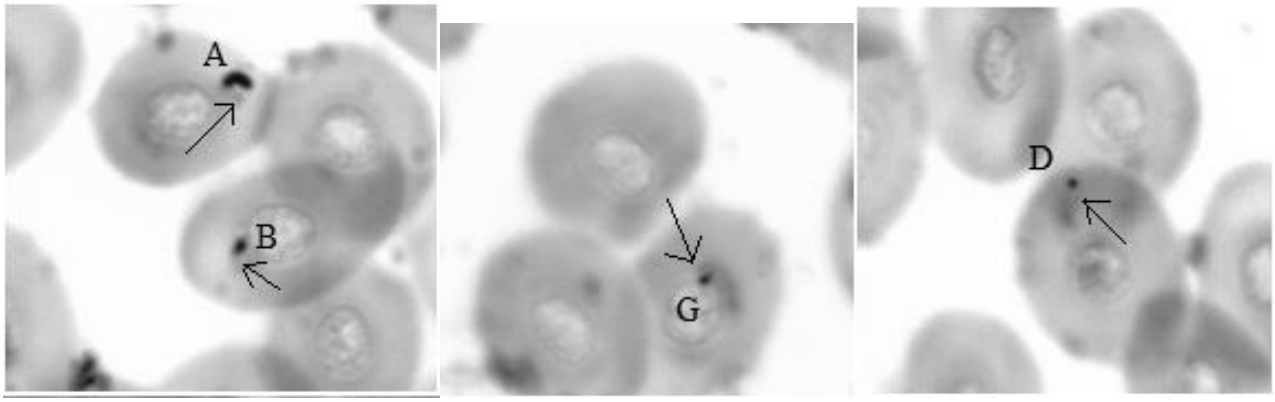

Figure 1. the Micronuclei (MNi) in blood of *C. punctatus*. A = Notched nuclei; B, G = lobed nuclei; D = micronuclei.

COD (r = 0.1516, t = 2.65). Change in pH can change the aspects of water chemistry, e.g. as pH increases (at Site 3), smaller amounts of ammonia is needed to reach a level that is toxic to fish. As pH decreases (at Site 2), the concentration of metals in water may increase because higher acidity increases their ability to be dissolved from sediments into the water and fishes were affected. Sim (2004) and Sujaul et al. (2012) have reported similar findings. The acidity of natural lake ecosystems due to high rich organic matter and metals were reported by Sujaul et al. (2012), Wetzel (1983) and Chapman (1988).

Temperature and dissolve oxygen (DO)

There are many factors that can influence the lake temperature. Water temperature can fluctuate seasonally, daily, and even hourly, especially in smaller sized streams or lake. Temperature affects the concentration of dissolved oxygen in water body. The dissolve oxygen in water is often attributed to the fact that the oxygen is dissolve more during the period of high catabolic activities by photosynthesis. DO decreases as turbidity, TDS and TSS increases (Joshi et al., 2009). The concentration of dissolve oxygen in lake is also affected by velocity of water, aquatic plants, TDS, TSS organic waste and urban human activities. In the present study, D.O. has strong significant negative correlation with BOD (r = 0.3459, t = 6.38), COD (r = 0.5854, t = 12.5), TSS (r = 0.3428, T = 6.32) and TDS (r = 0.3063, t = 5.57). Summer is usually the most crucial time for dissolve oxygen levels because stream flows tend to lessen and water temperature tend to increase. In the present study, the minimum DO concentration was observed in month of May-June at different sites when compared with months of July-August and November-December. In general, DO levels less than 3 mg/l are stressful to most aquatic organisms. Most fishes die at 1-2 mg/l. However, fish can move away from low DO areas. Water with low DO from 2-0.5 mg/l are considered hypoxic, waters with less than 0.5mg/l are anoxic.

Biological oxygen demand (BOD) and chemical oxygen demand (COD)

A quantitative relationship exists between the amount of oxygen required to convert a definite amount of any organic matter to CO_2, water and NH_3 (Joshi et al., 2009). BOD is an indication of the organic load and it is a pollution index especially for water bodies receiving organic effluents (Ndimele, 2012). There are significant negative correlation between BOD and TSS (r = 0.1557, t = 2.73)

and BOD and TDS (r = 0.1673, t = 2.93). Whereas significant positive correlation was observed in BOD and COD (r = 0.9984, t = 31.27). The value of BOD and COD were increased with increase in the pollution loads. Correlation analysis showed that COD had a significant negative relationship with pH (r = 0.1516, t = 2.65), DO (r = 0.5854, t = 12.5), TSS (r = 0.1815, t = 3.19) and TDS (r = 0.1905, t = 3.36).

Total suspended solids (TSS) and total dissolve solids (TDS)

The highest total suspended solids were recorded in month of May-June which vary from 303 ± 1.13 to 447 ± 1.24 mg/l. During July-August, it recorded from 213 ± 1.08 to 278 ± 1.10 mg/l and in November-December, it recorded from 184 ± 1.06 to 210 ± 1.13 mg/l. Total dissolve solids was recorded as 150.0 ± 1.12 to 186.75±1.20 mg/l during May-June. During July-August, it recorded from 187.32 ± 1.20 to 232.32 ± 1.34 mg/l. During November-December, was from 212.32 ± 1.21 to 265.34 ± 1.42 mg/l. Correlation analysis showed that TSS and TDS had a significant positive relationship (r = 0.9998, t = 86.58). With the above observation we concluded that due to the deposition of metals, pesticides residues and other organic wastages in the lake, pH of lake water become disturbed and it affects the vegetation in lake. Due to heavy deposition of organic waste and increase in concentration of ammonia, lake has eutrophication condition and this BOD level increase and DO level decrease, affect fish diversity and its health too. Similar results in biological aspect and physico-chemicals parameters were documented by several authors (Joshi et al., 2009; Sinha and Biswas, 2011; Shinde et al., 2011; Sujaul et al., 2012).

Micronucleus

Micronucleus (MN) test in fish erythrocytes of polluted water bodies were demonstrated to be a sensitive biomarker to detect genotoxic damage induced by complex mixture of contaminants in water. These complex mixtures of pollutants are responsible for multiple effects in aquatic organisms as it affect the functions of many organs, reproductive status, species survival, population size and ultimately biodiversity too (Bickham et al., 2000; Dixon et al., 2002). These carcinogenic and mutagenic compounds present in polluted water because of the discharge of industrial, agricultural and urban wastages may exert damage beyond that of individual and may be active through following generations. For ecotoxicological studies fishes are reported to be suitable organisms because they play different roles in the heterotrophic web, undergo bioaccumulation, and respond to mutagens at low concentration such as environmental pollutions (Cavas and Ergene-Gozukara, 2005; Kushwaha et al., 2012).

In field study, MN test is one of the most applicable techniques to identify genomic alterations in aquatic animals because this procedure is technically easier and more rapid than the microscopic analysis of chromosomal aberrations in metaphase (Fagr et al., 2008). Biomonitoring studies were carried out in native lake fish species *C. punctatus* at different sites. Consequently, maximum scoring in MN-frequency was observed in fish at Site 4 (fish received water from sewage waste) then Site 3 (fish received water from domestic water), Site 5 (fish collected from industrial effluent), Site 2 (fish received water from Metal workshop) and Site 1 (fish received water from agriculture run off). However, a number of studies applying the MN test in erythrocytes of fish in polluted water failed to reveal the impact of associated with well known genotoxic mixtures in polluted water such as heavy metals, pesticides or hydrocarbons. Many factors may be responsible for scoring of MN such as age, sex size, species, metabolic capacity, DNA repair efficiency and defense mechanisms, development of adaptive mechanisms of tolerances to chemical stress and rate of dead or damaged cells to maintain normal physiological conditions or inhibition of nuclear division which is required for MN expression (Rodriguez et al., 2003; Cavas and Ergene-Gozukara, 2005). Formation of MN occurred during anaphase from lagging acentric chromosome or chromatid fragments caused by misrepair of DNA breaks or unrepaired DNA breaks or at anaphase malsegregation of whole chromosomes which may also lead to MN formation. Malsegregation of whole chromosomes has resulted to the hypomethylation of repeat sequences in centromeric and pericentromeric DNA, defects in kinetochore proteins or assembly, dysfunctional spindle and defective anaphase checkpoint genes (Mateuca et al., 2006; Fenech, 2007). Malsegregation of chromosomes and formation of MN probably incorporated with inappropriate kinetochore protein assemble at centromere (Bull et al., 2008). Kinetochore proteins such as CENPA and CENPB are greatly affected by methylation of cytosine and histone also. Defects in kinetochore proteins leads to defects in microtubules interactions of chromatids during spindle formation and this mutation caused MN formation during Anaphase (Ieniet al., 2008). Other mechanisms that could lead to MN formation from acentric fragments Include simultaneous excision repair of damage or inappropriate base incorporated in DNA (e.g. uracil) that are in proximity and on opposite complementary DNA strands. Such simultaneous excition repair events, particularly if the gap-filling step is not completed, leads to DNA-double strands breaks and MN formation. This process can greatly enhance the blood MN assay DNA break or MN formation treated with cytosine arabinoside during G_1 phase of the cell cycle which inhibit the gap filling step of excision repair (Fenech and Crott 2002).

Several workers (Smith, 1990; Vigana et al., 2002; Rodriguez et al., 2003; Lemos et al., 2005) observed similar results in genotoxic patterns of polluted water in

other fish species.

Conclusion

The result of the present investigation on genotoxic potential of the polluted water of Maheshara Lake suggested a serious concern about its potential to damage aquatic organisms especially fish. However for safeguarding aquatic environments and organisms, several studies are needed to explore the biological consequences of DNA damage in fish due to polluted water of Maheshara Lake.

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