ISSN No:-2456-2165

Assessment of Heavy Metal Composition and Cytogenotoxic Risk Potential of Dumpsite Soil and Water Collected from Kogi State University Students Halls of Residence

[1]Onoja, A.O.*, [1]Egbeja, T. I., [1]Kadiri, J. U., [2]Edogbanya, P. R. O., [2]Oguche, J. U., [1]Alaji, P. O., [1]Onifade, D. D., [1]Osagiede, D. O. and [1]Ibrahim, H. G.
[1]Animal and Environmental Biology Department, Natural Sciences Faculty, Kogi State University, Anyigba, Nigeria
[2]Plant Science and Biotechnology Department, Natural Sciences Faculty, Kogi State University, Anyigba, Nigeria *Corresponding

Abstract:- Waste disposal is a vital environmental problem in both developed and developing countries. Nigeria generates an estimate of 29.78 x 10^9 kg waste annually which decomposes to produce toxicants with high concentrations which pose serious ecological and health risks to organisms. This research was carried out to evaluate the heavy metal composition as well as cytogenotoxic risk potential of soil and water samples collected from the halls of residence of Kogi State University, Nigeria. The concentration of heavy metals (Copper (Cu), Iron (Fe), Lead (Pb), Zinc (Zn), Nikel (Ni), Arsenic (As), Cadmium (Cd), and Chromium (Cr)) in soil samples were determined using Atomic Absorption Spectroscopy (AAS) and pollution index (PI) was assess by considering the combined contributions of all studied metals while cytogenotoxic potential of soil and water samples were assessed by using the *Allium cepa* test. The average concentration of metals in the dumpsite soils followed the trend Fe>Zn>Cu>Ni>Cr>Cd>Pb>As. Pollution index (PI) result revealed all dumpsite soil samples were within the range of moderate to strong pollution indicating potential risk to human health. The samples induced significant inhibition of *Allium cepa* root growth, decreased Mitotic index (MI) down the groups and induction of cell abnormalities compared to the control. The observed cytogenotoxic effects were assumed to be instigated by the heavy metals present in the samples. These results suggest that the investigated samples may be contaminated and may be of health risk to human population in case of exposure through dermal contact and direct ingestion. A serious and urgent introspection of these sites are therefore required by the stakeholders to mitigate this threat.

Keywords:- *Waste disposal, Soil, Heavy Metal, Pollution, Health Risk.*

I. INTRODUCTION

Pollution is an aspect of environmental degradation involving increase in the concentration of both biological and non-biological parameters above the environmental baseline levels which are environment determined (UNIFTPA, 2012). Nigeria generates an estimation of 29.78 x 10^9 kg household waste annually (Ojolo et al., 2004) that must eventually be landfilled around residential areas or open space not equipped with a containment system, these waste decompose to produce organic and inorganic toxicants and many unidentified metabolites which can remain in the environment for decades with high concentration capable of posing serious ecological and health risks to organisms including humans inhabiting adjacent area (Edwards, 2002; Bouhafs et al., 2009; Feng et al., 2013; Gamar et al 2017). These pollutants are dangerous because they can bioaccumulate and biomagnify thereby interfering with the living tissues biochemistry processes (Goyer, 1991; Alloway and Ayres, 1995; Sawyer et al., 2006).

Soil and water are important environmental media that sustain life on earth, provides mechanical anchorage and favourable tilt serving as a reservoir of nutrients; they act as connecting links between organic, inorganic and living systems (Watanabe and Hirayama, 2001; Fred, 2012). However, in recent decades, soil and water are increasingly becoming sinks due to exposure to large quantity of hazardous contaminants through several activities including natural and anthropogenic which ultimately result to their contamination. These pollutants has prospects to pose severe risk to human health through numerous route of exposure which may include direct ingestion, consumption of contaminated groundwater, dermal contact, agricultural crops/food produce as well as food web/chain in the process of bioaccumulation and biomagnification (Kanmony, 2009). The influence of these pollutants affects man and also causes severe toxicity to various wildlife (Edwards, 2002; Feng et al., 2013), hence the need for environmental monitoring using Bioindicators which project numerous types of unique evidence such as: (I)

ISSN No:-2456-2165

primary cautioning of environmental damage; (II) primary cautioning of likely damage to human health on the basis of wildlife responses to pollution; (III) response interactions between exposed individual organisms to pollution and the effects amongst the population level; (IV) the combined influence of a range of environmental stresses on organisms, the population, community and ecosystem; and (V) the effectiveness of the corrective efforts in decontaminating soil and water-channels (Villela IV 2006).

Soil and water are predisposed to pollutant such as heavy metal, organic and microbial contamination due to anthropogenic activities resulting in bio-diversity change, decrease population size and soil and water communities overall activities (Ashraf and Ali, 2007). Therefore, ecological risks concern due to accumulation of heavy metals in the soil and water which compromises the ecosystem integrity and consequently calls for more ecological risk assessment (Islam et al., 2017). Heavy metal food contamination is hazardous and can influence toxic effects on health of an ecosystem as well as humans (Balkhair and Ashraf, 2016; Alghobar and Suresha, 2017). For instance heavy metal such as Cadmium in processing of Zinc among others is intensively used in the industries for rechargeable batteries, paints, stabilization of plastic, these can accumulates in the environment and gets to human diet through biomagnification causing cancer, reproductive dysfunction, hepatic and renal toxicity (Meyer, 1983; Alimba et al., 2012; Jaishankar et al., 2014).

The incidence of cancer disease amongst inhabitants of Niagara, United States exposed to chemicals discharged out of Love canal dumpsites (Janerich et al., 1981) and occurrence of low birth weight amongst inhabitants exposed landfill leachate hazardous chemicals (Vianna and Polan, 1984), amplified the consciousness about the linked between human health and continuous exposure to toxicants from dumpsites. This research was carried out to assess the cytogenotoxic potentials of dumpsite soil and water collected from the students' hall of residence, because at the moment, there is no sanitary dumpsite within the University, making leachate-toxicant movement through soil and water (groundwater and surface water) highly possible. Therefore, information on the degree of pollution in the study area is required for setting background values in monitoring pollutants build-up and assessing the quantities of pollutants translocation through the ecosystem compartments.

II. MATERIAL AND METHODS

➢ *Study Area*

Anyigba is located between longitude 7º 12ʹ East of the Greenwich meridian and latitude 7º 36ʹ North of the Equator in Dekina LGA Kogi State Nigeria, characterized by both rainy and dry seasons with about 747mm mean annual rainfall and average maximum and minimum temperature of 33.2ºC and 22.8ºC respectively. The students halls of residence admits about 12, 000 population and is characterized with high students activities on campus (Figure 1).

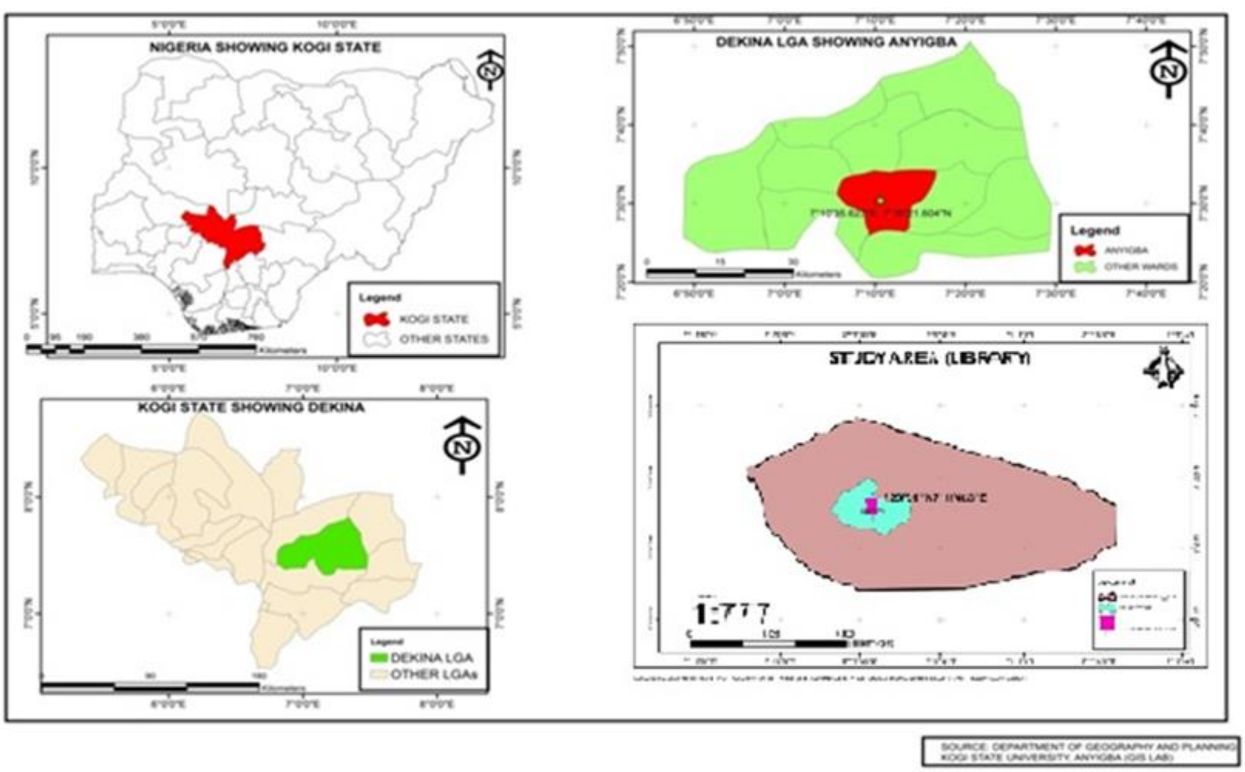

Fig 1

- *Apparatus Sanitisation and Chemicals used*

The used apparatus includes; *Allium cepa*, beakers, Polythene bags, petri dishes, coplin jar and dissecting set. Detergent solution was used to wash the beakers and petri dishes and rinsed with distilled water. Acetone was used to clean the stainless steel tools in the dissecting set. The digestion chemicals/reagents used for this study are HNO_3, $HCLO_4$ and HF (Sigma-Aldrich, Germany) while Giemsa Stain, Acetocarmine, Carnoy Fixative, NHCL were used for *Allium* root tip preparation.

- *Soil and Water Sample Collection and Preparation*

Samples of dumpsite soil and water were collected from the Halls of Residence within the campus of the Kogi State University, Anyigba, Nigeria. The soil samples were obtained between the depths of 0 to 15cm depth, at intervals of about 20m apart from four sites. The soil samples collected from a sampling site were prepared into a compound sample to make a single sample per site. Each pooled samples were properly air-dried in a storeroom to evade cross impurity, mixed and then sieved with a 0.5 mm size sieve to remove unwanted particles. The sieved soil was ground to fine powder. Fifty grammes (50g) of each soil sample were selected and kept in a labelled polythene bags and taken to IITA (International Institute of Tropical Agriculture), Ibadan, Nigeria for its subsequent digestion and heavy metal analysis. The remaining portion were extracted according to Cabrera and Rodriquez, (1999) as sampled soil is suspended in distilled water (ratio 1:2 w/v) and stirred at interval for 24 hours and filtered thereafter. The filtered extracts were considered as 100%. Also, water samples were collected from four different water sources within the hall of residence. It was collected into small covered bucket of five (5) litres each from the water sources (borehole) and brought into the laboratory for further investigation. Distilled water was used as negative control.

- *Digestion and determination of heavy metal in soil sample*

Miroslav and Vladimir (2005) procedure was used to leach the heavy metals from soil samples with modifications. Two grammes (2g) of the fifty (50g) each of dried and sieved selected soil samples were accurately weighed into rinsed beaker and concealed using rinse glass material; 30ml 1:1 of HNO_3: deionized water was added to each beaker containing soil and gently boiled using hot plate in the hood with constant stirring at interval till the volume decreased or condensed to about five (5) ml. Then ten (10) ml 1:1 HNO_3: deionized water added again and the process repeated to obtain another five (5) ml which were diluted properly in standard flasks and kept preserved at 4^0C before Atomic Absorption Spectrometer (AAS) was used for heavy metal analysis for Copper (Cu), Iron (Fe), Lead (Pb), Zinc (Zn), Arsenic (As), Cadmium (Cd), Nikel (Ni) and Chromium (Cr). All chemicals used were of standard grade (Sigma-Aldrich, Germany).

- *Risk Assessment Indices*

Pollution index (PI) was used assess the level of pollution within the study area. The PI assesses the degree of pollution by computing the combined influence of all polluting soil metals average concentration with the permissible level in accordance to formula given by Lee, et al., 1998.

$$\text{Pollution index (PI)} = \frac{1}{n}\left(\frac{\text{M1}}{\text{TL1}} + \frac{\text{M2}}{\text{TL2}} \dots\dots\dots\dots\dots\dots . \frac{\text{Mn}}{\text{TLn}}\right)$$

Where n represents sum total of metals M1, M2….., Mn are the average concentration of the polluting metals while TL1, TL2 ….., TLn are the permissible concentration of each polluting metals.

- *Allium cepa Preparation*

Fresh small commercial *Allium cepa* of the same size weighing about 3-3.5g were obtained from Anyigba main market, Kogi State, Nigeria. The loose outer scales of the bulbs and brownish bottom plates were scrapped with razor leaving the root primordial ring intact for the emergence of new root and rinsed with clean water, equal millitres of the samples were measured into 18 beakers properly labelled with three beakers for each samples, which means the bulbs were divided into three replicas, the bulbs were then placed directly on the beaker containing investigation extracts and kept in a dark place, the soil and water samples under investigation were divided into three portions and successfully applied to the *Allium* roots every 24 hours. So each 24 hours, the roots obtained fresh bath of the sample solution.

- *Cytological Experimental Procedure*

The experiment was terminated after 72 hours and the roots were removed with a forceps and observed macroscopically for related parameters (shape of the roots, number, colour and turgescence). The lengths of each root from each bulb/the whole root bundle were measured using a meter rule giving one value for each bulb. The root tips were harvested and then placed in Carnoy fixative (glacial acetic: ethanol 1:3) for 24 hours. The root tips are hydrolysed for 10 minutes with 10% of hydrochloric acid to soften it after properly washed with distilled water for 3 minutes. Then the root tips were placed on a clean microscope slide and squashed in two drops of Acetocarmine until a turbid suspension achieved and covered with cover slip. Excess stain was drained off by the use of tissue paper and gently applying pressure. The slides were mounted and observed under light microscope using 1000× magnification and the various mitotic stages per treatment were recorded. About 100 cells/ root tip dividing cells were scored.

- *Cytological Data Analysis*

Numbers of dividing cells (prophase, metaphase, anaphase and telophase) were taken per investigated samples for total number of cells. Experiment was conducted in triplicate each. Mitotic Index (MI) for the cells was computed by determining the mitotic cell frequency at the root tip cells as:

$$\text{Mitotic Index (MI)} = \frac{\text{Total number of dividing cell}}{\text{Total number of cell examined}} \text{X } 100$$

Data collected in this study were subjected to Analysis of Variance (ANOVA). Values were recorded as mean ± Standard Error of Mean (SEM); $P < 0.05$ was used to measure significant; Duncan Multiple Range Test (DMRT) was used to separate means where significant. All computations were done using Statistical Package for Social Sciences (SPSS) version 23.

III. RESULTS AND DISCUSSION

➢ *Heavy metals analysis of soil samples*

Heavy metals in soils have health risk potentials when investigated metals concentrations possibly leaked into the environment when situations such as acidic precipitation prevail in the study area. The mean concentration levels o heavy metals Copper (Cu), Iron (Fe), Lead (Pb), Zinc (Zn), Arsenic (As), Cadmium (Cd), Nikel (Ni) and Chromium (Cr) in each soil sampled from the vicinity are contained in Table 1. The average concentration of metals in the sampled soils followed the trend Fe>Zn>Cu>Ni>Cr>Cd>Pd>As. The level of Iron (Fe) found in the soil was within the WHO standard limit, the concentration of Zinc (Zn) and Copper (Cu) exceeds the standard limit of WHO significantly, while the other metals were below the tolerated limit (Table 1).

Heavy metals are hazardous and might have negative influence on the health of human as a result of their accumulation properties (Tressou, et al., 2004). The zinc values were higher than the WHO standard, indicating high zinc-metal-contamination in the studied environment. Excessive zinc intake has been reported to be toxic to human and causes pancreatic degradation, anaemia, reproductive defects, developmental defects, nausea, electrolyte imbalance and lethargy (Prasad, 1984; Domingo, 1994). Copper concentrations were much higher than the WHO-permitted level. Copper toxicity is reported to be linked with reactive oxygen species (ROS) when the cells storage capacity for surplus copper has been exceeded (Linder, 2001). Excessive copper intake may cause dermatitis, neurological disorders and liver cirrhosis (Fairweather-Tait, 1988). The arsenic values were lower than the WHO standard. However, persistent or chronic exposure to ingestion of inorganic arsenic might induce blackfoot disease and to inorganic arsenic results in some health problems such as the skin, liver, respiratory tract, gastrointestinal tract, hematopoietic system, cardiovascular system and nervous system (Chen et al., 2001; Mandal and Suzuki, 2002). Cadmium concentrations were lower but uptake of 1.5-9 mg/day is toxic and injurious to humans. It could impair the kidneys function and indicates chronic toxicity such as hepatic dysfunction, cancer growth/tumours, poor reproductive capacity and hypertension (Waalkes, 2000). The WHO classified chromium as a human carcinogen and a number of scholars have revealed that Chromium (VI) compounds have potentials to upsurge lung cancer risk (Ishikawa et al., 1994; Takahashi et al., 2005). Lead has the capacity to affect human health by causing liver damage and renal failure (Emmerson, 1973). Nickel is a ferromagnetic elements occurring in soil naturally and a recognized environmental pollutant, entering natural water channels through waste water because treatment processes poorly removed it (Cain and Pafford, 1981), with skin allergic reaction in human as the most adverse health effect (Das et al., 2008). Nickel is recognized as carcinogens, genotoxic, nephrotoxic, hepatotoxic, haematotoxic, immunotoxic, neurotoxic, reproductive toxic and pulmonary toxic. There is a need for watchfulness on anthropogenic sway connected with the dumpsites which accepts every types of multifarious wastes (Aboyade, 2004; Ogundiran and Afolabi, 2008), because factors responsible for heavy metals concentration in dumpsite soil may comprise of: (1) the form of wastes discarded (2) the waste pollutant stabilisation ability to adequate permissive limits before discarding (3) the wastes time frame within the dumpsite.

Heavy metal (ppm)	SS 1	SS 2	SS 3	SS 4	WHO LIMIT
Fe	293.781±29.675	76.680±50.261	6.479±29.675	253.880±29.675	295000
Zn	101.647±57.828	33.898±57.828	64.111±57.828	104.976±57.828	50
Cu	80.120±6.174	60.839±6.174	65.895±6.174	87.479±6.174	36
Pb	0.6386±0.1410	0.1737±0.2832	0.452±0.1410	0.7092±0.1410	85
As	0.250±0.040	0.021±0.030	0.084±0.030	0.265±0.030	0.39
Cd	0.655±0.362	0.575±0.362	0.399±0.362	1.045±0.362	0.8
Ni	4.447±1.790	1.186±0.722	1.239±0.722	4.349±0.722	35
Cr	2.165±0.042	1.508±0.402	1.348±0.402	1.755±1.229	100

Table 1: -AAS Heavy metals Concentrations in soil samples within the dumpsites (Mean of triplicate ± Standard Error of Mean)
SS: soil sample

- *Risk Assessment*

Pollution index (PI) was used to evaluate the level of pollution within the study area. The result of the index used to envisage the degree of pollution in the soils is presented in Table 2 by considering the combined contributions of all studied metals. The results of this index showed that all sampled soils were within moderate to strong pollution range. This indicates that pollution level in the studied area is becoming an issue because of the potential harm of these pollutants to human health. Monitoring and assessment of the environment have become a necessity in discovering where insidious pollution occurred and the sources as well as kind of pollutants involved (McBee and Bickham, 1990; Propst et al., 1999).

Sample ID	Pollution index (PI)
SS 1	0.7
SS 2	0.9
SS 3	1.17
SS 4	2.93
Interpretation: PI>1=polluted, PI<1=unpolluted	

Table 2:- Pollution index for dumpsites soil

- *General Toxicity, Root Growth Inhibition and Cytological Effects of Allium cepa*

During harvest turgescence and hardness, turgidity and slackness, brownish colourations and root tip bending were observed in some roots as a sign of toxicity. Some showed little or no growth as compared to the controlled water sample which showed longer roots and lesser general toxicity p<0.05. Soil sample 4 showed high level of root growth inhibition based on the fact that there was no root growth indicating high levels of toxins present (Table 3 and Figure 2).

A remarkable decline of mitotic index (MI) in all groups was identified compared to the control at P<0.05 using DMRT, a considerable frequency of aberrations was also identified in root mitotic cells grown in the investigated samples. The most encountered were disturbed spindles and laggard chromosomes when compared with the control. Aside these, the prophase stage of division were seen more than the other phases; all the values in other phases (metaphase, anaphase and telophase) were reduced compared to the control, probably due to the toxicants interference with the cell cycle and mitotic activities resulting in cycle inhibition, since mitotic inhibition is recognised as sign of cytotoxic effect (Olorunfemi et al., 2011). These may be indications that the samples had considerable toxic level (Table 3 and Figure 3) because decreases of mitotic index below 50% (cytotoxic limit value) in comparison to control usually have sublethal effects (Panda and Sahu, 1985; Andrade et al., 2008). Studies have reported various forms *Allium* and animal chromosomal and mitotic abnormalities observed in polluted/contaminated soils from different parts of the world (Rank and Neilson, 1997; Alimba et al., 2012; Masood and Malik, 2013; Souza et al., 2013). Aberrant analysis allowed estimation of genotoxic effects, clastogenic and aneugenic actions of investigated samples because chromosome aberrations could result from spindle formation inhibition as well as DNA synthesis blockage.

Treatments	Root length	Prophase	Metaphase	Anaphase	Telophase	Mitotic Index	Laggard	Disturbed Spindle
NC	0.91 ± 1.10	10.00^a	10.33^c	16.33^c	14.00^c	$54.00±12.49^c$	$0.00±0.00^a$	$0.33±0.58^a$
SS 1	0.00± 0.00*	28.33c	1.33a	1.33a	1.00a	32.00±5.00 a	1.50±0.70ab	$1.00±0.00^a$
SS 2	1.06 ± 0.25	17.33ab	3.67ab	2.67a	5.00a	28.67±4.62a	1.50±0.58ab	$1.25±0.96^a$
SS 3	0.61 ±0.30	21.67bc	4.00b	3.33a	5.00a	33.67±1.53ab	2.00±1.00b	$1.33±0.58^a$
SS 4	NG	--	--	--	--	--	--	--
WS 1	0.08 ± 0.08	13.67a	3.00ab	3.67a	5.00a	25.33±5.86a	2.33±1.53b	$1.33±0.58^a$
WS 2	0.79 ± 0.06	24.00bc	4.33b	8.33b	9.33b	46.00±9.54bc	1.67±0.58ab	$1.33±0.58^a$

Table 3:- Effect of various investigated dumpsite soils and water samples on the root length, and Cytological Effects of *Allium cepa* (*Mean of triplicate ± Standard Error of Mean*)

SS: Soil Sample, WS: Water Sample, NC: Negative Control, NG: No Growth, ^{a,b,c}: means on the same row with different superscript differ significantly at P<0.05, * significant value different from negative control at P<0.05.

ISSN No:-2456-2165

Fig 2A:- *Allium cepa* planting on investigated soil extract

Fig 2B:- Root emergence after 72h

Fig 2C:- Root length after 72h

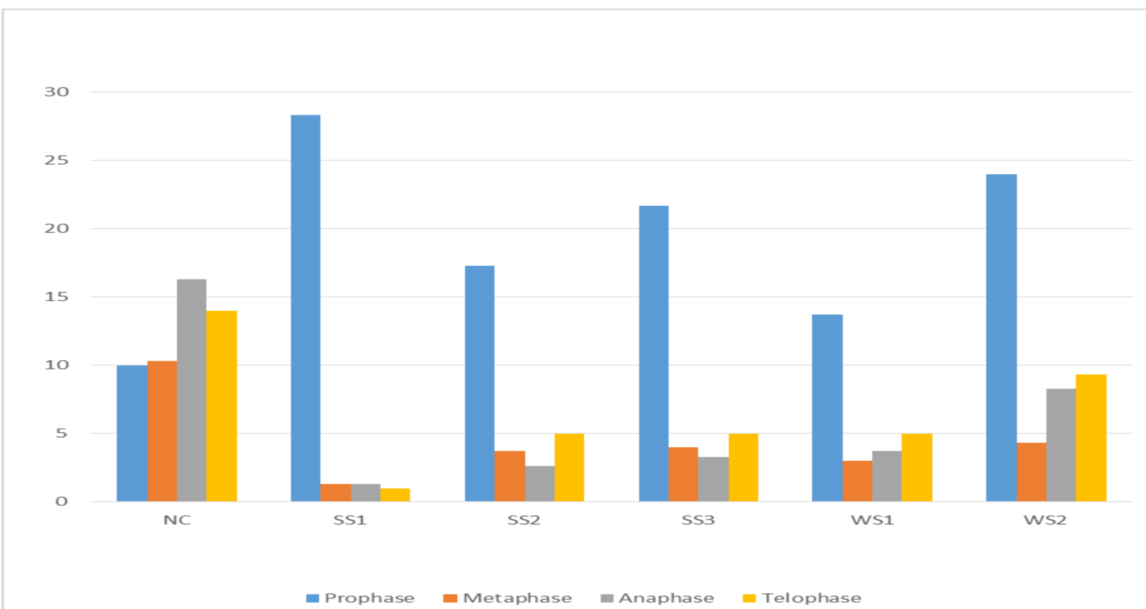

Fig 3:- Chart showing Mitotic phase Index of investigated soil and water samples.

IV. CONCLUSION

The composition of heavy metal (Iron, Zinc, Copper, Lead, Arsenic, Cadmium, Nikel and Chromium) and cytogenotoxic risk of dumpsites soil and water within the students' hall of residence was assessed for the degree of contamination from the dumpsites. The concentration of all the metals and the pollution index clearly indicates anthropogenic effect due to more number of students' admitted annually and that the studied area have been polluted to a certain degrees which causes undue potential damage to organisms health including humans living in the location, if not now, in future by their possibility to diffused through to the soil to underground waters recycled for use on campus. The study also submits that reaction of *Allium cepa* root length and genetic materials can be a yardstick to estimate the effects of possible cyto-genotoxic constituents in the environs since pollutants at non-toxic level could be found toxic at certain level by bioaccumulation and Biomagnification making it to constitute a danger to health. It also supports or enhances knowledge about soil and water contaminations in a comprehensive way in order to avoid possible dangers connected with non-degradable contaminants or pollutants which can continue to accumulates and interfere with the biomolecules in the living tissues, thus posing serious risk to human health. From this study dumping of wastes on these sites had been for decades, therefore the need to regulate and possibly urgently evacuate dumpsites from these halls of residence as well as awareness of the pollution status needs to be created especially among students and university community.

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