

## ENVIRONMENTAL HEALTH RISK ASSESSMENT OF HOSPITAL WASTEWATER IN ENUGU URBAN, NIGERIA

EZE C.T.<sup>1</sup> and ONWURAH I.N.E.<sup>2</sup>

<sup>1</sup> Environmental pollution & Toxicology Unit, Department of Biochemistry, Federal University Oye-Ekiti, P.M.B. 373, Oye, Ekiti State, Nigeria, <sup>2</sup> Pollution Control and Biotechnology Unit, Department of Biochemistry, University of Nigeria, Nsukka, Enugu State, Nigeria  
E-mail: chukwuebukaeze@ymail.com

### ABSTRACT

An important hydrogeologic problem in areas of high faults formations is high environmental health hazard occasioned by microbial and heavy metals contamination of ground waters. Consequently, we examined the microbial load and heavy metals concentration of hospital wastewater discharged into the environment at Park Lane General Hospital Enugu Urban, Nigeria. The microbial counts, characteristics and frequency of occurrences of the isolated microorganisms over a given period of time were determined by cultural, morphological and biochemical characteristics using established procedure while the varying concentrations of the identified heavy metals were determined using spectrophotometric method. The microbiological analyses showed a mean total aerobic bacteria counts from  $13.7 \pm 0.65 \times 10^7$  to  $22.8 \pm 1.14 \times 10^{10}$  cfu/ml, mean total anaerobic bacteria counts from  $6.0 \pm 1.6 \times 10^3$  to  $1.7 \pm 0.41 \times 10^4$  cfu/ml and mean total fungal counts from  $0 \pm 0$  to  $2.3 \pm 0.16 \times 10^5$  cfu/ml. The isolated micro-organisms which included both pathogenic and non-pathogenic organisms were *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Bacillus subtilis*, *Proteus vulgaris*, *Klesbsiella pneumonia* and *bacteriodes sp.* The only fungal isolate was *Candida albican*. The heavy metals identified in the leachate were Arsenic, Cadmium, Lead, Mercury and Chromium and their concentrations ranged from  $0.003 \pm 0.00082$  to  $0.14 \pm 0.0082$  mg/l. These values were above WHO permissible limits while others fall within the limits. Therefore hospital waste water can pose environmental health risk when not properly treated before discharge, especially in geologic formations with high fault formations.

**Keywords:** Hospital wastewater, microbial counts, bacterial isolates, fungal isolates and heavy metals

### 1. Introduction

Wastewater is referred to as any water whose quality has been adversely abused by anthropogenic influence (Ekhaize and Omavwoya, 2008). This includes liquid waste discharged from hospitals, domestic homes and industries, agricultural and commercial sectors among others. The world is faced with problems related to the management of wastewater. This is due to extensive industrialization, increasing population density and high urbanized societies (EPA, 1993; McCasland et al., 2008). The effluents generated from hospital activities contribute to the natural environmental pollution load. This is a great burden in terms of wastewater management and can consequently lead to a point-source pollution problem, which not only increases treatment cost considerably, but also introduces a wide range of chemical pollutants and microbial contaminants to the environment (EPA, 1993, 1996; Eikelboom and Draaijer, 1999; Amir et al., 2004). The presence of high concentrations of these pollutants above the critical values stipulated by national and international regulatory bodies is considered unacceptable in receiving environment. This is because, apart from causing a major drawback in wastewater treatment systems, they also lead to eutrophication and various health impacts in humans and animals (CDC, 2002; Runion, 2008). In recent years, the reuse of treated effluent that is normally discharged to the environment from wastewater treatment plants is receiving an increasing attention as a reliable water resource. In many countries, wastewater treatment for reuse is an

important dimension of water resources planning and implementation. This is aimed at releasing high quality water supplies for potable use. Some countries, such as Jordan and Saudi Arabia have national policies to reuse all treated wastewater effluents, thus have made considerable progress towards this end. In China, sewage use in agriculture has developed rapidly several decades ago and millions of hectares are irrigated with sewage effluent. The general acceptance is that wastewater use in agriculture is justified on agronomic and economic grounds, although care must be taken to minimize adverse health and environmental impacts (FAO, 1992; Metcalf and Eddy, 2003; Rietveld et al., 2009; Sowers, 2009). Furthermore, wastewater reuse is increasingly becoming important for supplementing drinking water needs in some countries around the world. The option of reuse of wastewater is becoming necessary and possible as a result of increased climate change, thus leading to droughts and water scarcity, and the fact that wastewater effluent discharge regulations have become stricter leading to a better water quality (Rietveld et al., 2009).

The aim and objectives of the study was to assess the environmental health risk associated with hospital wastewater by determining the varying concentrations of the heavy metal constituents as well as to isolate, identify and characterize the associated microbial population.

## **2. Microbiological characteristics of wastewater**

The major microorganisms found in wastewater are viruses, bacteria, fungi, protozoa and helminthes. Although various microorganisms in water are considered to be critical factors in contributing to numerous waterborne outbreaks, they play many beneficial roles in wastewater influents (Kris, 2007). Traditionally, microorganisms are used in the secondary treatment of wastewater to remove dissolved organic matter. The microbes are used in fixed film systems, suspended film systems or lagoon systems, depending on the preference of the treatment plant. Their presence during the different treatment phases can enhance the degradation of solids, resulting in less sludge production (Ward-Paige et al., 2005a). Wastewater microbes are also involved in nutrient recycling, such as phosphate, nitrogen and heavy metals. If nutrients that are trapped in dead materials are not broken down by microbes, they will never become available to help sustain the life of other organisms. Microorganisms are also responsible for the detoxification of acid mine drainage and other toxins in wastewater (Ward-Paige et al., 2005b). Microbial pollutants can also serve as indicators of water quality. The detection, isolation and identification of the different types of microbial pollutants in wastewater are always difficult, expensive and time consuming. To avoid this, indicator organisms are always used to determine the relative risk of the possible presence of a particular pathogen in wastewater (Paillard et al., 2005). For instance, enteric bacteria, such as coliforms, *Escherichia coli*, and faecal streptococci are used as indicators of faecal contamination in water sources (DWAf 1996; Momba and Mfenyana, 2005). To indicate viral contamination, bacteriophages (somatic and F-RNA coliphages) are used. Also, *Clostridium perfringens*, a faecal spore-forming bacterium, which is known to live longer in the environment and reported to be resistant to chlorine, is used as an indicator for the presence of viruses, protozoa or even helminthes eggs (Payment and Franco, 1993; Grabow et al., 1997).

## **3. Heavy metal characteristics of wastewater**

The heavy metals of importance in wastewater treatment are Arsenic, Cadmium, Chromium, Cobalt, Copper, Iron, Lead, Mercury, Silver and Zinc among others. Living organisms require varying amounts of some of these metals (Calcium, Cobalt, Chromium, Copper, and Zinc among others) as nutrients (macro or micro) for proper growth. Other metals (Silver, Cadmium, Gold, Lead and Mercury among others) have no biological role and hence are non-essential (Metcalf and Eddy, 2003; Hussein et al., 2005). Heavy metals are one of the most persistent pollutants in wastewater. Unlike organic pollutants, they cannot be degraded, but accumulate throughout the food chain, producing potential human health risks and ecological disturbances. Their presence in wastewater is due to discharges from hospitals, residential dwellings, groundwater infiltration, and industrial discharges (Hussein et al., 2005; Silvia et al., 2006). The danger of heavy and trace metal pollutants in water lies in two aspects of their impact. Firstly, heavy metals have the ability to persist in natural ecosystems for an extended period, and, secondly, they have the ability to accumulate in successive levels of the biological food chain (Fuggle, 1983). Although heavy

metals are naturally present in small quantities in all aquatic environments, it is almost exclusively through human activities that these levels are increased to toxic levels (Nelson and Campbell, 1991). The methods for determining the concentrations of these metals vary in complexity according to the interfering substances that may be present. Typical methods of determining their concentrations include flame atomic absorption, electro-thermal atomic absorption, inductively coupled plasma, and inductively coupled plasma (ICP)/mass spectrometry (APHA, 2001).

#### **4. Materials and method**

**Sample Collection:** The wastewater samples were collected from three (3) wastewater outlets with pre-cleaned sterile and dried containers at around 11am on Thursday-10/05/2012 from three (3) different wards at Parklane General Hospital Enugu Urban. The three (3) sampling points were designated wws1 - wws3. Wastewater from Medical Ward-(wws1), wastewater from New Born Baby ward-(wws2) and wastewater from Surgical Ward-(wws3). The samples were transported to the laboratory where it was preserved under laboratory conditions and analysis initiated within 3hours of sample collection.

**Digestion of Wastewater Sample:** The EPA vigorous digestion method described by Gregg (1989) was adopted. 100ml of each of the representative wastewater samples were transferred into Pyrex beakers containing 10ml of concentrated HNO<sub>3</sub>. The samples were boiled slowly and then evaporated on a hot plate to the lowest possible volume (about 20ml). The beakers were allowed to cool and another 5ml of Conc. HNO<sub>3</sub> was added. Heating was continued with the addition of Conc. HNO<sub>3</sub> as necessary until digestion was complete. The samples were evaporated again to dryness (but not baked) and the beakers were cooled, followed by the addition of 5ml of HCl solution (1:1 v/v). The solutions were then warmed and 5ml of 5M NaOH was added, then filtered. The filtrates were transferred to 100ml volumetric flasks and diluted to the mark with distilled water. These solutions were then used for Microbial and heavy metal analysis.

**Microbial Analysis:** All the microbial isolates were identified by standard microbiological techniques. The bacteria were characterized using cultural identification, Gram staining reaction and other biochemical tests including: Catalase, Starch hydrolysis, Coagulase, Indole, Citrate, Motility, Oxidase, Urease and Sugar fermentation tests according to Bergey's Manual of Determinative Bacteriology (9th edition). The fungal isolates were identified using their growth rate, colony morphology and microscopic morphological features.

**Heavy Toxic Metal Analysis:** The varying concentrations of the identified heavy toxic metals were determined by atomic absorption spectrophotometry (model S Series, S4 AAS system) (APHA, 2001).

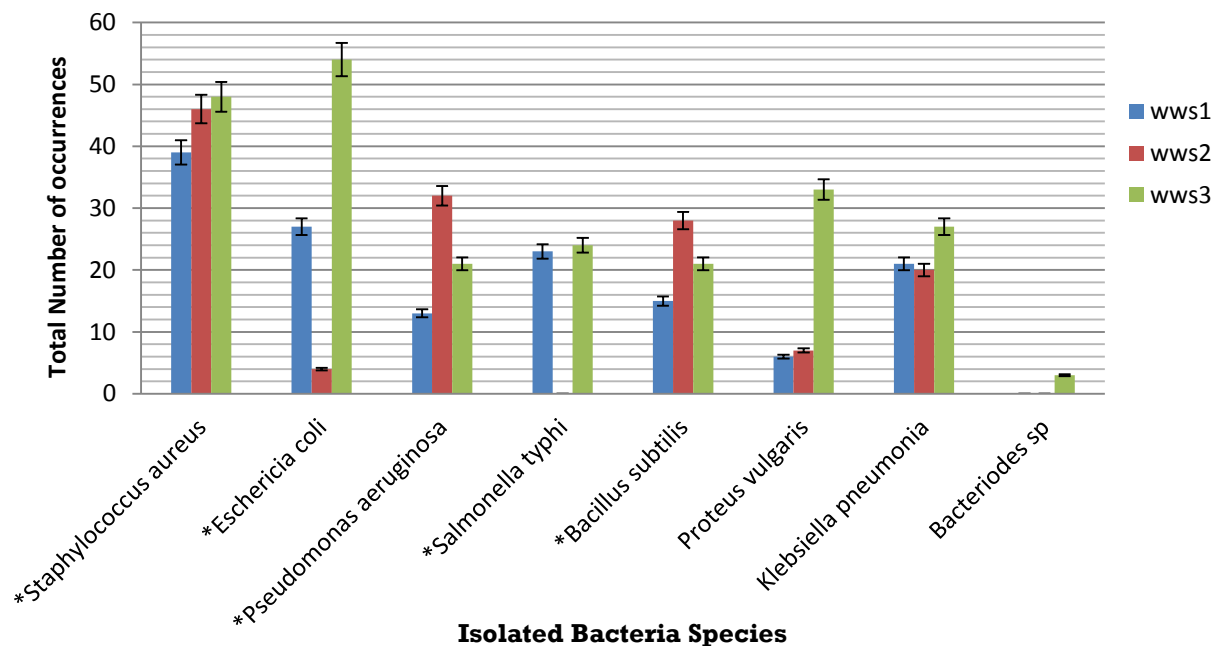
#### **5. Results and discussion**

The result of characterization and identification of pure microbial isolates in the hospital wastewater samples reveals that on Tryptone Soya Agar (TSA) *Pseudomonas aeruginosa* appeared greenish-blue; *Staphylococcus aureus* appeared deep yellow; *Escherichia coli* formed mucoid white colonies; *Bacillus subtilis* formed large dry and flat dirty brown colonies; *Proteus vulgaris* formed large milk coloured colonies; *Salmonella typhi* formed white colonies with black centers, while *Klebsiella* sp appeared bright pink and *Bacteriodes* appeared as large brown flat colonies.

On Sabourand dextrose Agar (SDA), fungal yeast cell appeared as smooth large elevated whitish-brown colonies. This is an indication of the presence of only *Candida albican*-a pathogenic fungus.

The identified micro-organisms revealed in this study included both pathogenic and non-pathogenic organisms. According to Oyeleke and Istifanu (2009), pathogenic organisms refer to organisms which pose great harm to the public and environmental health. The result for the frequency of occurrences of the isolated bacterial species showed high frequency distribution of *staphylococcus aureus* in the wastewater samples (Fig.1). The wastewater sample from the surgical ward (wws3) showed a highest frequency of occurrence of both *Staphylococcus aureus* and *Escherichia coli* (Fig.1). The presence of these organisms portends potential risk to nearby farms and water bodies, which may be contaminated by wastewater effluent. The occurrences of

the pathogenic organisms revealed in this study were in agreement with the report of Giroletti and Lodola (1993). The result also revealed the presence of *Candida albicans*-a pathogenic fungus as the only isolated fungi specie in the hospital wastewater samples though wastewater from the new born baby ward (wws2) showed no fungal occurrence (Fig.2). This might suggest that hospital wastewater does not provide a favorable media for the growth of fungi. The mean total bacteria counts ranged from 103 - 1010 mg/l while that of fungal counts ranged between 0 -105 (Table 1) and fell within the range reported by other workers (Ferando et al., 1994; Ingham et al., 1989).



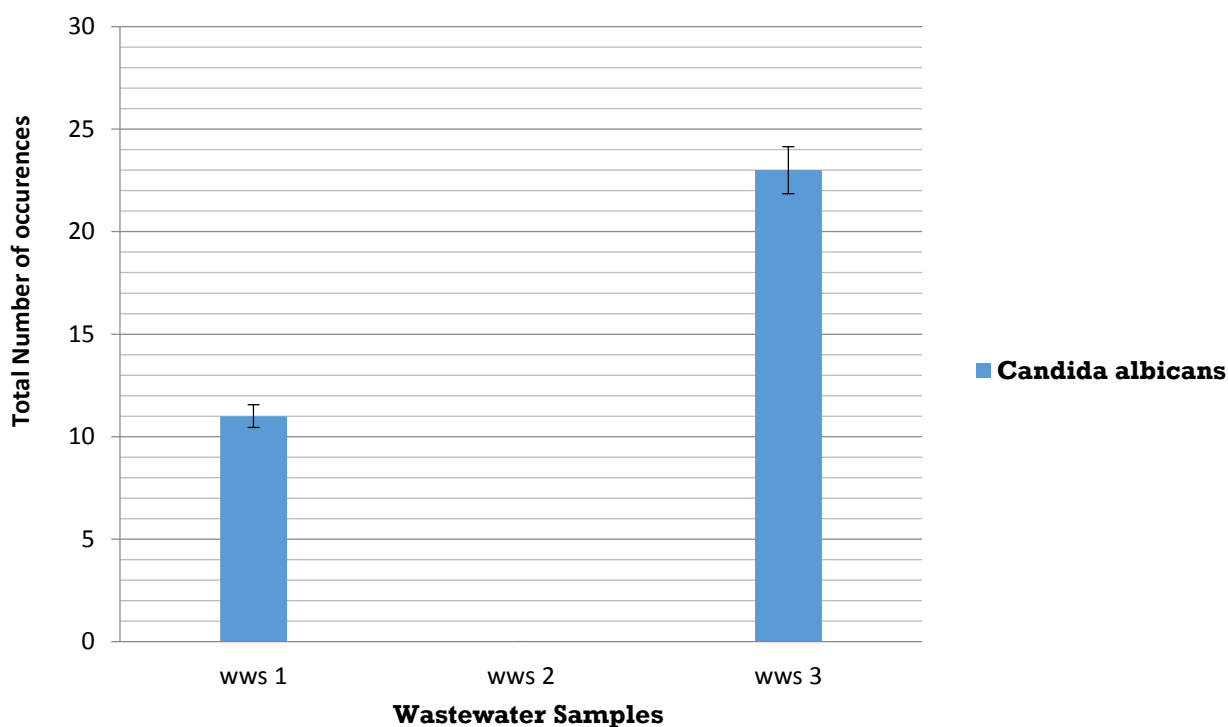
**Figure 1:** Result for the Frequency of Occurrences of Bacteria Isolates from hospital wastewater samples. wws1:-sample from point 1, wws2:-sample from point 2 and wws3:-sample from point 3. The pathogenic isolates are marked with (\*).

**Table 1:** Biochemical Characteristics of different bacteria Isolates

Bacteria	Gram stain	Motility	Indole	Starch hydrolysis	Catalase	Coagulase	Urea	Oxidase	Lactose	Citrate
<i>Bacillus subtilis</i>	+	+	-	+	+	-	-	+	-	-
<i>Pseudomonas aeruginosa</i>	-	+	-	-	-	-	-	+	-	-
<i>Staphylococcus aureus</i>	+	-	-	-	+	+	-	-	+	-
<i>Escherichia coli</i>	-	+	+	-	-	-	-	-	+	-
<i>Bacteriodes spp</i>	-	+	-	-	-	-	+	-	-	+
<i>Klebsiella pneumonia</i>	-	-	-	-	-	-	+	-	+	+
<i>Salmonella typhi</i>	-	+	-	-	-	-	-	-	-	-
<i>Proteus vulgaris</i>	-	+	+	-	-	-	+	-	-	+

+ = Positive reaction  
 = Negative reaction

The mean total bacteria counts were generally higher than those of fungal, irrespective of sampling locations. The presence of high coliform densities in the wastewater samples is an indication of faecal pollution of the environment. The introduction of wastewater in the environment brings about increased amount of organic matter and essential nutrients which can influence changes in the micro flora and micro fauna. Aluyi et al., (2006) noted that high bacteria counts reflected the level of pollution in the environment which is an indication of organic matter present. When evaluating the effects of hospital wastes on microbial communities, it is important to note that target organisms vary between hospital wastes. Indigenous communities of bacterial and fungal populations are very complex and they have the important task of cycling nutrients. Although the result for the test of significant difference between the varying concentrations of the identified heavy metals in the hospital wastewater and WHO permissible limits showed an insignificant variation ( $P > 0.05$ ), the result as well revealed some degree of variations among the samples with respect to WHO permissible limits (Table 3).



**Figure 2:** Result for the Frequency of Occurrences of *Candida albicans* in the hospital wastewater samples. wws1:-sample from point 1, wws2:-sample from point 2 and wws3:-sample from point 3.

**Table 2:** Result of Mean Microbial Counts in the Hospital Wastewater Samples

Mean Microbial counts	WWS <sub>1</sub>	WWS <sub>2</sub>	WWS <sub>3</sub>
Mean Total Aerobic Bacteria Count (cfu/ml)	$14.4 \pm 0.86 \times 10^{10}$	$13.7 \pm 0.65 \times 10^7$	$22.8 \pm 1.14 \times 10^{10}$
Mean Total Anaerobic Bacteria Count (cfu/ml)	$6.0 \pm 1.6 \times 10^3$	$4.0 \times 10^3 \pm 2.5 \times 10^3$	$1.7 \pm 0.41 \times 10^4$
Mean Total Fungal Count (cfu/ml)	$1.1 \pm 0.82 \times 10^3$	Nil	$2.3 \pm 0.16 \times 10^5$

wws<sub>1</sub>:-sample from the medical ward  
 wws<sub>2</sub>:-sample from the new born baby ward  
 wws<sub>3</sub>:-sample from the surgical ward  
 cfu/ml:-colony forming unit per milliliter

**Table 3:** Result of Mean Concentrations of Heavy Toxic Metals in the Hospital Wastewater Samples.

Heavy Toxic Metals	WWS <sub>1</sub> conc. (mg/l)	WWS <sub>2</sub> conc. (mg/l)	WWS <sub>3</sub> conc. (mg/l)	WHO Limit (mg/l)
Arsenic (As)	0.08 ± 0.0082	0.06± 0.0163	0.14 ± 0.0082	0.1
Cadmium (Cd)	0.13 ± 0.0163	0.04 ± 0.0163	0.05 ± 0.0163	0.1
Lead (Pb)	0.083± 0.0082	0.02 ± 0.0082	0.06 ± 0.0082	0.1
Mercury (Hg)	0.012± 0.00082	0.007± 0.00163	0.009 ± 0.0082	0.01
Chromium (cr <sup>+6</sup> )	0.09 ± 0.0163	0.003± 0.00082	0.012 ± 0.0052	0.01

wws<sub>1</sub>:-sample from the medical ward

wws<sub>2</sub>:-sample from the new born baby ward

wws<sub>3</sub>:-sample from the surgical ward

mg/l:- milligram per liter

## 6. Conclusions

Conclusively, it was observed that hospital wastewater have negative influence on the environment. The frequency of occurrences of the microbial isolates, high mean microbial counts and the varying concentrations of heavy toxic metals obtained in this study suggests that hospital wastewater in the environment is a major health and environmental threat, which therefore calls for a proper regulatory system on the disposal of hospital wastewater especially in the developing countries. Since large amounts of wastewater effluents are passed through sewage treatment systems on daily basis, it is therefore advised that there is a need to remedy and diminish the overall impacts of these effluents on the receiving environment. In order to comply with wastewater legislations and guidelines, wastewater must be treated before discharge. This can be achieved through the application of appropriate treatment processes, which will help to minimize the risks to public health and the environment. To achieve unpolluted wastewater discharge into the environment, there is the need for careful planning, adequate and suitable treatment, regular monitoring and appropriate legislation. This will enhance science-based decisions and ensure the sustainability of the environment and the health of plants and animals. There is also a need to ensure that effluents standards and limitations, as set by regulatory bodies are not compromised.

## REFERENCES

1. Aluyi A.S.A., Ekhaise O.F. and Adelusi, M.D (2006). Effect of human activities and oil pollution on the microbiological and physiological quality of Udu River, Warri, Nigeria. *Journal of Applied of Sciences*. 6(5): 1214-1219.
2. Amir H.M, Ali R.M, Farham K (2004). Nitrogen removal from wastewater in a continuous flow sequencing batch reactor. *Pak. J. Biol. Sci.* 7(11): 1880-1883.
3. APHA (2001). *Standard Methods for the Examination of Water and Wastewater*, 20th edition. APHA, Washington D.C.
4. *Bergey's Manual of Determinative Bacteriology*. 9th edition, williams & wilkins, Batimore ISBN 0-683-00603-7
5. CDC (2002). U.S. Toxicity of Heavy Metals and Radionucleotides. Department of Health and Human Services, Centers for Disease Control and Prevention. Savannah river-site health effects subcommittee (SRSHES) meeting: Available from [http://www.cdc.gov/nceh/radiation/savannah/SRSHES\\_Toxicity\\_jan02.htm](http://www.cdc.gov/nceh/radiation/savannah/SRSHES_Toxicity_jan02.htm). Accessed 07/12/2007.
6. DWAF (1996). *South Africa Water Quality Guidelines for Domestic Use*, 2nd edition, Pretoria, South Africa.
7. Eikelboom D.H and Draaijer A (1999). Activated sludge information system (ASIS). Available from <http://www.asissludge.com>. Accessed 09/11/2007.
8. Ekhaise F.O and Omavwoya B.P (2008). Microbiological Influence of Hospital wastewaters discharged into the environment of University of Benin Teaching Hospital. *American-Eurasian J. Agric. & Environ. Sci.*, 4 (4): 484-488.
9. EPA (1993). *Constructed wetlands for wastewater treatment and wildlife habitat*. Available from <http://www.epa.gov/owow/wetlands/construct>. Accessed 14/01/2008

10. EPA (1996). U.S. Environmental Protection Agency, American Society of Civil Engineers, and American Water Works Association. Technology Transfer Handbook: Management of Water Treatment Plant Residuals. EPA/625/R-95/008. Washington DC.
11. FAO (1992). Wastewater treatment and use in agriculture. Food and Agriculture Organization (FAO) of the United Nations Irrigation and Drainage Paper 47. Available from <http://www.fao.org/..docrep/T0551E/T0551E00.htm>. Accessed 10/11/2010.
12. Ferando, A and Heen, C. (1994): Hospital waste: An environmental hazard and its management. NBRI, Lucknow. 14
13. Fuggle R.F (1983). Nature and Ethics of Environmental Concerns. In: Environmental Concerns in South Africa. Fuggle RF, Rabie MA (eds), Juta Cape Town.
14. Giroletti E and Lodola L (1993). ISPRA COURSES: Waste Treatment and Management. Medical Waste Treatment.1-8
15. Grabow W.K, Very A, Uys M and Villiers J.C (1997). Evaluation of the application of bacteriophages as indicators of water quality. South Africa Water Research Commission, Final Report 540/1/98.
16. Gregg, L.W. (1989): Water analysis Handbook. H.A.C.H Company, USA. Pp. 33 – 39.
17. Hussein H, Farag S, Kandil K and Moawad H (2005). Tolerance and uptake of heavy metals by Pseudomonads. Process Biochem. 40: 955-961.
18. Ingham, S.,Rhodes, G., G. Huys, J. Swings, P. McGann, M. Hiney, P. Smith and Pickup, W.R. (1989). Distribution of oxytetracycline resistance plasmids between aeromonads in hospital and aquaculture environments: Implications of Tn172 in dissemination of the tetracycline resistance determinant Tet A. Appl. Environ. Microbiol. 66(9): 3883-3890.
19. Kris M (2007). Wastewater Pollution in China. Available from <http://www.dbc.uci.wsu.stain/suscoasts/krismin.html>. Accessed 16/06/2008.
20. McCasland M, Trautmann N, Porter K and Wagenet R (2008). Nitrate: Health effects in drinking water. Available from <http://pmep.cee.comell.edu/facts.slides-self/facts/nit-heefgrw85.html>. Accessed 05/04/2008.
21. Metcalf X and Eddy X (2003). Wastewater Engineering: Treatment and Reuse. In: Wastewater Engineering, Treatment, Disposal and Reuse. Tchobanoglous G, Burton FL, Stensel HD (eds), Tata McGraw- Hill Publishing Company Limited, 4th edition. New Delhi, India.
22. Momba M.N.B and Mfenyana C (2005). Inadequate treatment of wastewater: A source of coliform bacteria in receiving surface water bodies in developing countries- Case Study: Eastern Cape Province of South Africa. In: Water Encyclopedia- Domestic, Municipal and Industrial Water Supply and Waste Disposal. Lehr JH, Keeley J (eds). John Wiley & Sons Inc, pp. 661-667.
23. Nelson W.O and Campbell P.G.C (1991). The effect of acidification on the geochemistry of Al, Cd, Pb and Hg in freshwater environments: a literature review. Environ. Pollut. 71: 91-130.
24. Onyeleke, S.B and Istifanus, N (2009): The microbiological effects of hospital wastes on the environment. African Journal of Biotechnology. 8(7):1253-1257.
25. Paillard D, Dubois V, Thiebaut R, Nathier F, Hogland E, Caumette P and Quentine C (2005). Occurrence of *Listeria* spp. In effluents of French urban wastewater treatment plants. Appl. Environ. Microbiol. 71(11): 7562-7566.
26. Payment P and Franco E (1993). Clostridium perfringens and somatic coliphages as indicators of the efficiency of drinking water treatment for viruses and protozoa cysts. Appl. Environ. Microbiol. 59: 2418- 2424.
27. Ratsak CH, Maarsen KA and Kooijman SAL (1996). Effects of protozoa on carbon mineralization in activated sludge. Water Res. 30(1): 1-12.
28. Rietveld L.C, Meijer L, Smeets P.W.M.H and Hoek J.P (2009). Assessment of Cryptosporidium in wastewater reuse for drinking water purposes: a case study for the city of Amsterdam, The Netherlands. Water SA, 35(2): 211-215
29. Runion R (2008). Factors to consider in wastewater-2. Ezine Articles (April 14). Available from <http://ezinearticles.com/?factors-To-Consider-Wastewater--2&id=1108477>. Accessed 09/05/2008.
30. Silvia D, Ana M and Juan C.G (2006). Evaluation of heavy metal acute toxicity and bioaccumulation in soil ciliated protozoa. Environ. Int. 32(6): 711-717.
31. Sowers AD (2009). The effect of wastewater effluent on fish and amphibians species. A Doctoral Dissertation presented to the Graduate School of Clemson University.
32. Ward-Paige C.A, Risk M.J and Sherwood O.A (2005a). Reconstruction of nitrogen sources on coral reefs:  $\delta^{15}N$  and  $\delta^{13}C$  in gorgonians from Florida Reef Tract. Mar. Ecol. Prog. Ser. 296: 155-163.
33. Ward-Paige C.A, Risk M.J, Sherwood O.A and Jaap W.C (2005b). Clonid sponge surveys on the Florida Reef Tract suggest land-based nutrient inputs. Mar. Pollut. Bull. 51: 570-570.
34. WHO (1997). World Health Organization Permissible Limits. Environmental Health Criteria 2nd edn No. 54. Geneva, Switzerland.