



Journal of Environmental Science and Technology

ISSN 1994-7887

science
alert

ANSI*net*
an open access publisher
<http://ansinet.com>

Investigation of Toxicity of Detergents

T. Yahaya, J. Okpuzor and E.O. Oladele

Department of Cell Biology and Genetics, University of Lagos, Nigeria

*Corresponding Author: T. Yahaya, Department of Cell Biology and Genetics, University of Lagos, Nigeria
Tel: +2348033550788*

ABSTRACT

The general health problems of detergents with emphasis on the weight, hematology and histopathology were investigated by ingesting 20 mice (about 30 days old) with detergent solutions. The mice were grouped into five sets, comprising of five mice each. The control group (group 1) was given ordinary water while groups 2-5 were given 25, 50, 75 and 100% v/v of the detergent solutions, respectively as the only sources of water. The mice had access to feed (F.A Feeds, Lagos) *ad libitum* in suspended, meshed-bottom cages for about 4 weeks. The Relative Growth Rate (RGR) in percentage of the control mice was 7.1 while the RGR of the mice exposed to 25, 50, 75 and 100% v/v of the detergent solutions were -33.2, -43.9, -52.5 and -59.3, respectively. A significant difference ($p < 0.05$) exists between the mean weight of the control mice and the mean weights of the mice placed on the different concentrations of the detergent. The hematology analysis of the exposed mice showed marked reduction in the packed cell volume, hemoglobin, white blood cells, red blood cells and total protein. Furthermore, the blood of the exposed mice had dose-dependent hypochromasia, macrocytosis, microcytosis, eosinophilia and anisocytosis. A significant difference ($p < 0.05$) also exists between the mean blood parameters of the control mice and the mean blood parameters of the mice exposed to the various concentrations of the detergent. The internal organs (heart, kidney, liver and reproductive organs) of the exposed mice showed marked histological changes compared to the control mice and the extent of the injuries increased with the concentrations of the detergent solutions. The results of the research highlight health hazards of detergent exposure and underscore the needs for proper handling and disposal of detergents.

Key words: Mice, toxicity, detergent, hematology, histopathology

INTRODUCTION

Detergents are materials which aid in the removal of dirt or other foreign matters from contaminated surfaces. Until the 1940s, soap was the only important detergent in the market but today, soap is one of the numerous detergent products. According to one ancient Roman legend, soap derives its name from Mount 'Sapo' in Italy where animals were sacrificed to appease the gods. The mixture of the melted animal fats and wood ashes that ran down to the clay soil near Tiber River at Mount 'Sapo' was found to make washing easier than ordinary water for women inhabiting the area (Ronni, 2004). But another report from a medical document Papyrus, said that soap-making dates back to about 1500BC when a combination of animal vegetable oils and alkaline salt was used to form a soap-like material. However, modern technology has led to the creation of synthetic detergents that have gradually replaced soaps in the laundry. The first detergent was used chiefly for hand dish-washing and fine fabric laundering. This was followed by the development of multi-purpose laundering detergent introduced in the United States of

America in 1946. Today, detergent market is a highly competitive one where different brands compete with one another to get customers (Khurana, 2002).

The chemical components of detergents are often called surface-active agents or surfactants because they act upon surfaces. A common feature of detergents is that they are made up of comparatively large molecules (Molecular weight over 200 g). One part of the molecules is soluble in organic material and the other part is soluble in water. Before, synthetic detergents were made by treating an aromatic or benzene-type compound with sulfuric acid, followed by neutralization with alkali to convert the product to its sodium salt. These detergents, however, became a public nuisance because they were neither soluble nor biodegradable. The aromatic compound was later replaced with a so-called linear alkyl-type compound in the process described above and the detergent produced was as effective as the former kind in its detergent action and was more biodegradable and soluble (Redmond, 2011).

Detergents are widely used in both industrial and domestic premises to wash cloths, equipment, installations, heavy-duty machines and vehicles. They are also useful in pesticide formulations for agricultural purposes and dispersing of spills at sea (Lightowers, 2004; Chun and You, 2009). However, the increasing domestic health hazards and environmental pollution arising from detergent exposure are reducing the popularity of its usefulness (Ezemonye and Enuneku, 2005). But most of the health hazards are caused by dearth of information on the toxicity of detergents which makes people to handle or dispose detergents carelessly. Therefore, a thorough investigation on the health risks and ecological disorders that may arise from improper handling and disposal of detergents needs to be done. This has become necessary because presently, there is no alternative to detergent. Worldwide, governments are making concerted efforts to keep environments free of pollutants through policies, environmental conferences and conventions. In Nigeria, Federal Government is emphasizing the needs for adequate human and environmental protections in any technological and socio-economic development by strictly asking industrial operators to control the effects of their wastes (DPR, 1991). But in Nigeria and other countries, only few chemicals have been ecologically tested for safety despite their alleged environmental and ecological impacts (Ogundiran *et al.*, 2010). There is no doubt that detergent exposure posed health risks to man and other animals but the doubt lies in what it actually does to them and how dangerous it really is (Easy Essay, 2007).

Although some researchers have described the effects of detergents on animals and plants, most of the works were done on the basis of spirometry or radiology, or both. An in-depth analysis of the effects of detergent exposure on different organs and systems in human or animal has not been published. Therefore, the present study was aimed at investigating the toxicity and general health problems that may arise from detergent exposure.

MATERIALS AND METHODS

Test substance: The detergent used in this study was collected from a dumping ground of a detergent factory in Ibadan, Nigeria. To avoid contaminations, the detergent was collected the same day it was dumped. The chemical constituents of the detergent as stated by the manufacturers include surfactants, sodium sulfate, sodium carbonate, optical brighteners, phosphate, poly-carboxylates and enzymes.

Animal husbandry: Swiss mice (about 30 days old) of either sex, bred in the animal house University of Lagos, Nigeria, were used for the research. They were housed in suspended,

meshed-bottom cages and were left for about seven days to acclimatize under continuous light (12 h light/darkness) before the commencement of the research. Pelletized feeds from FA Feeds, Lagos, were given to the mice *ad libitum*.

Experimentation: Four concentrations (25, 50, 75 and 100% v/v) of the detergent solutions were prepared. The mice were grouped into five sets of 5 mice each after taken their initial weights. The control mice were resided in cage 1 and were given ordinary water. The exposed mice in cages 2, 3, 4 and 5 were given 25, 50, 75 and 100% v/v of the detergent solutions, respectively as the only sources of water. The experiment commenced in mid-October, 2010, a week after the purchase of the mice and was allowed to stay for about four weeks. At the end of the 4th week, the final weights of the mice were taken and their blood and internal organs (liver, kidney, heart and lung) were examined in the laboratory.

Calculation of Relative Growth Rate (RGR): The relative growth rates of the mice were calculated using the formula below:

$$\text{RGR (\%)} = \frac{W_f - W_i}{T} \times 100$$

Where:

W_f = Final weight

W_i = Initial weight

T = Period of exposure

Hematological examination: The mice were sedated with chloroform in the laboratory. Total death was prevented to allow continuous flow of blood for proper blood collection. Each mouse was pegged down on a work bench and held firmly with office pins. Surgical blades were used to cut through the chest region of the mice in a dorsal-ventral direction. The blood was then collected from a beating heart using a Na Heparinized capillary tube through capillary action into EDTA bottles. EDTA serves as anti-coagulant and so also the Na heparin in the capillary tube. The blood parameters (PCV, HB, WBC and RBC) were determined using Sysmex auto-analyzer while the total protein was determined using VET 360 Veterinary Refractometer (Phoenix series).

Preparation of internal organs for histopathology examinations: The internal organs (heart, kidney, liver and reproductive organs) of the mice were prepared for histopathology examinations using the method of Taylor *et al.* (2003).

Statistical analysis: A database file was created in a personal computer and all statistical analysis was carried out with the Statistical Package for Social Sciences (SPSS) version 17 for windows and Microsoft Office Excel 2007. Comparison of data among exposed and control groups were calculated using Student's t-test. $p < 0.05$ was considered statistically significant.

RESULTS

Table 1 showed that the chemical constituents of Bonux detergents had adverse effects on the weights of the mice. For example, the mean weight change of the control mice was 1.9 g while the

mean weights change of the mice that were placed on 25, 50, 75 and 100% v/v solutions of the detergent were -9.3, -12.3, -14.7 and -16.6 g, respectively. Also, the relative growth rate of the control mice in percentage was 7.1 while the relative growth rates of the mice exposed to 25, 50, 75 and 100% v/v of the detergent solutions were -33.2, -43.9, -52.5 and -59.3, respectively.

The results of the hematology examinations of the exposed mice showed marked reduction in the blood parameters. The mean PCV change of the control mice was 0.3% whereas the mean PCV lost by the mice placed on 25, 50, 75 and 100% v/v of the detergent solutions were -8.0, -11.9, -17.0 and -21.4%, respectively (Table 2). From table 3, the mean change in the HB of the control mice was 0.1 g dL⁻¹ while the mean HB change of the mice exposed to 25, 50, 75 and 100% v/v of the detergent solutions were -2.4, -3.8, -6.3 and -9.1 g dL⁻¹, respectively. Furthermore, the control mice showed mean WBC increase of 34 mm³, whereas the mice treated with 25, 50, 75 and 100% v/v of the detergent solutions showed mean WBC increases of 151, 186, 262 and 548 mm³, respectively (Table 4). From Table 5, the mean RBC change was 0.1×10¹² while the mean RBC lost by the mice exposed to 25, 50, 75 and 100% v/v of the detergent solutions were -0.5, -0.8, -1.4 and -2.7×10¹², respectively. And finally, the mean total protein change of the control mice at the end of the exposure was 0.08 g dL⁻¹ while the mean total protein lost by the mice placed on 25, 50, 75 and 100% v/v of the detergent solutions were 0.7, 1.2, 2.6 and 3.2 g dL⁻¹, respectively (Table 6). Generally, the mice placed on 25 and 50% v/v of the detergent solutions showed slight microcytosis, macrocytosis, eosinophilia and ariscytosis while the mice placed on 75 and 100% v/v of the detergent solutions showed chronic conditions of the above diseases.

The results of the histopathology analysis of the internal organs of the exposed mice showed marked histological changes. The heart showed cell swelling, interstitial edema and loss of cellular outline, hyalinization of tissues and atrophy of cells in 25, 50, 75 and 100% v/v of the detergent solutions, respectively. The kidney had cortical necrosis and tubular atrophy in the 25, 50 and 75% v/v of the detergent solutions and severe necrosis in the 100% v/v of the solution. The liver

Table 1: Effects of the detergent solutions on the weights of the mice

Detergent solutions	Minimum weight	Maximum weight	0 Day mean weight	4th week mean weight	Mean weight change	Relative growth rate	p-values
Ordinary water (control)	27.8	32.3	28.8±0.78 ^a	30.65±1.16 ^b	1.9	7.1	0.022*
25%	18.5	29.6	28.7±0.81 ^a	19.40±0.89 ^b	-9.3	-33.2	<0.0001*
50%	14.1	29.6	28.6±0.72 ^a	16.30±1.38 ^b	-12.3	-43.9	<0.0001*
75%	13.1	29.8	28.8±0.93 ^a	14.10±0.73 ^b	-14.7	-52.5	<0.0001*
100%	11.1	29.5	28.6±0.87 ^a	12.00±0.66 ^b	-16.6	-59.3	<0.0001*

Data were expressed as Mean±SD: When *p<0.05: Significant from control and when **p>0.05: Not significant from control Mean values in the same row with different superscripts were significantly different at p<0.05

Table 2: Effects of the detergent solutions on the PCV (%) of the mice

Detergent solutions	Minimum PCV	Maximum PCV	Mean PCV at 0 day	Mean PCV at 4th week	Mean PCV change	p-values
Ordinary water (control)	36.8	38.4	37.5±0.57 ^a	37.80±1.19 ^a	0.3	0.4529**
25%	28.5	38.3	37.9±0.38 ^a	29.90±1.20 ^b	-8.0	<0.0001*
50%	25.2	38.4	37.9±0.43 ^a	26.00±0.61 ^b	-11.9	<0.0001*
75%	19.6	39.3	38.1±0.88 ^a	21.10±1.06 ^b	-17.0	<0.0001*
100%	15.8	39.1	38.1±0.85 ^a	16.70±0.59 ^b	-21.4	<0.0001*

Data were expressed as Mean±SD: When *p<0.05: Significant from control and when **p>0.05: Not significant from control Mean values in the same row with different superscripts were significantly different at p<0.05

Table 3: Effects of the detergent solutions on the HB (g dL⁻¹) of the mice

Detergent solution	Minimum HB	Maximum HB	Mean HB at 0 day	Mean HB at 4th week	Mean HB change	p-values
Ordinary water (control)	12.5	13.2	12.8±0.26 ^a	12.90±0.22 ^a	0.1	0.6151**
25%	9.9	13.3	12.8±0.41 ^a	10.40±0.83 ^b	-2.4	0.0002*
50%	9.1	13.4	13.0±0.31 ^a	9.18±0.23 ^b	-3.8	<0.0001*
75%	6.0	13.2	12.8±0.31 ^a	6.50±0.45 ^b	-6.3	<0.0001*
100%	4.0	13.6	13.2±0.37 ^a	4.10±0.31 ^b	-9.1	<0.0001*

Data were expressed as Mean±S; When *p<0.05: Significant from control and when **p>0.05: Not significant from control Mean values in the same row with different superscripts were significantly different at p<0.05

Table 4: Effects of the detergent solutions on the WBC of the mice

Detergent solution	Minimum WBC	Maximum WBC	Mean WBC at 0 day	Mean WBC at 4th week	Mean WBC change	p-values
Ordinary water (control)	3.850	4.600	4.140±230 ^a	4.174±272 ^a	34	0.8367**
25%	4.210	4.750	4.152±317 ^a	4.303±285 ^a	151	0.4845**
50%	4.100	4.890	4.044±434 ^a	4.230±470 ^a	186	0.8231**
75%	3.600	4.630	4.100±366 ^a	4.362±242 ^a	262	0.2795**
100%	3.840	5.220	4.358±250 ^a	4.906±300 ^a	548	0.0081*

Data were expressed as Mean± SD: When *p<0.05: Significant from control and when **p>0.05: Not significant from control Mean values in the same row with different superscripts were significantly different at p<0.05

Table 5: Effects of the detergent solutions of the RBC of the mice

Detergent solution	Minimum RBC	Maximum RBC	Mean RBC at 0 day	Mean RBC at 4th week	Mean RBC change	p-values
Ordinary water (control)	4.8	6.1	5.40±0.57 ^a	5.48±0.52 ^a	-0.1	0.8223**
25%	4.1	6.1	5.26±0.65 ^a	4.86±0.71 ^a	-0.5	0.1528**
50%	4.1	6.3	5.38±0.66 ^a	4.58±0.59 ^a	-0.8	0.03313*
75%	3.2	6.0	5.34± 0.56 ^a	3.90±0.59 ^b	-1.4	0.0020*
100%	2.5	6.3	5.60±0.62 ^a	2.90±0.29 ^b	-2.7	<0.0001*

Data were expressed as Mean±SD: When *p<0.05: Significant from control and When **p>0.05: Not significant from control Mean values in the same row with different superscripts were significantly different at p<0.05

Table 6: Effects of the detergent solution on the total protein of the mice

Detergent solution	Minimum total protein	Maximum total protein	Mean total protein at 0 day	Mean total protein at 4th week	Mean total protein change	p-values
Ordinary water (control)	6.5	7.0	6.86±0.23 ^a	6.94±0.11 ^a	0.08	0.5061**
25%	6.0	7.2	6.82±0.31 ^a	6.08±0.20 ^b	0.70	<0.0001*
50%	5.0	7.3	6.90±0.20 ^a	5.70±0.50 ^b	1.20	0.0002*
75%	3.9	7.1	6.90±0.30 ^a	4.30±0.40 ^b	2.60	<0.0001*
100%	3.1	7.2	6.90±0.20 ^a	3.70±0.50 ^b	3.20	<0.0001*

Data were expressed as Mean±SD: When *p<0.05: Significant from control and when **p>0.05: Not significant from control Mean values in the same row with different superscripts were significantly different at p<0.05

suffered cellular edema in the 25% v/v, partial necrosis in the 50 and 75% v/v solutions and partial hepatic necrosis in the 100% v/v solution. The lungs of the exposed mice showed interstitial edema in the 25% v/v solution, tissue edema in the 50% v/v solution and severe necrosis and cellular atrophy in the 75 and 100% v/v solutions. The reproductive organs (ovary and testis) of the exposed mice showed fat deposits and degenerations. No significant findings were observed in the internal

organs of the control. Generally, the exposed mice showed eye problems, tumors on the body, sluggishness and high mortality rate compared to the control.

DISCUSSION

The reduction in the growth rates of the exposed mice supports the findings of Esenowo and Ugwumba (2010) who reported that sub-lethal concentrations of a detergent in an aquarium tank reduce the weights of catfish (*Claris gariepinu*) exposed. Rejeki *et al.* (2006), also observed that chronic concentrations of detergent LAS retarded the growth of the sea bass larvae exposed significantly. However, the result contradicts Faremi and Oloyede (2010) who did not observe any significant growth reduction in albino rats exposed to detergent. The growth reduction observed in this study might have resulted from detoxification (Ojo and Oso, 2009), poor physiological conditions of the exposed mice which led to loss of appetite, sluggishness and high metabolic demand (Ogunbileje and Akinosun, 2011). Sodium silicate, a major component of detergents, has also been fingered in gastro-intestinal irritation leading to nausea, vomiting, diarrhea, severe digestive tract burns and weight loss in exposed animal (Warne and Schiffko, 1999).

The marked reduction in the blood parameters (PCV, HB, WBC, RBC and Total Protein) of the exposed mice compared to the control mice confirms the earlier report of Wadaan and Mubarak (2009) who reported blood parameters reduction in rabbit exposed to Sodium Dodecyl Sulfate (surfactant). The toxic effect of the anionic detergent (surfactant) has also been reported by Dehelean *et al.* (2004), to cause reduction in blood parameters and weight loss. The reduction in the blood parameters was as a result of destruction of the cells of the mice by the surfactant and traces amount of heavy metals found in detergents (Abulude *et al.*, 2007). Riaz *et al.* (2009) reported that blood and skin of an animal may be adversely affected by soaps and detergents used as anti-bacteria and insecticides because they do so by killing the cells of the germs and insects. The increase (although insignificant) in the White Blood Cells (WBC) of the exposed mice was as a result of immune response of the mice to the toxic components of the detergent solutions.

The fact that the heart, lung, kidney, liver and reproductive organs of the exposed mice showed marked histopathological changes confirms that the detergent solutions are pathogenic to mice. The histopathology changes were as a result of cytotoxic agents (Ayandiran *et al.*, 2009) in the detergent solutions which caused multi-organ injuries. This result supports the findings of Gholami *et al.* (2010) and Ogundiran *et al.* (2009, 2010) who reported that ingestion of detergent solutions can cause multi-organ damage leading to nervous system disorders, respiratory illness, birth defects, cancer and even death. Furthermore, the result is in line with Akanji *et al.* (1993) and Uaboi-Egbenni *et al.* (2009) both reported pathological damage in animal and plant exposed to detergent, respectively. However, this study did not find such diseases like cancer and asthma which have been suspected by some researchers (Kassem, 2010), probably due to short duration of the study. The sluggishness, low fecundity and high mortality rate observed in the exposed mice were direct consequences of the impaired health and poor physiological conditions of the mice.

The observed diseases call for careful handling and disposal of detergent. Alternatively, lipases such as microbial lipase can be introduced in to detergent during production to reduce the mass of toxic materials found in detergents (Hasan *et al.*, 2010).

CONCLUSION

The results of the research have shown that the detergent is toxic to the mice exposed. It is presumed that all other animals including man will suffer similar fate if exposed to the detergent solutions. Ordinarily, a matured man will not ingest detergent except by accident but children especially toddlers who are not conscious of their actions are the endangered population. Therefore,

extra care should be taken to monitor the activities of our toddlers and they should not be left alone. Detergents should be kept out of the reach of children and spilled detergent should be packed immediately. Finally, it is hoped that the results of this investigation will enlighten people on health risks posed by detergent exposure.

REFERENCES

- Abulude, F.O., M.O. Ogunkoya, R.F. Ogunleye, O. Emidun and A.I. Abulude, 2007. Assessment of the content of Pb, Cd, Ni and Cr in soaps and detergents from Akure, Nigeria. *Res. J. Environ. Toxicol.*, 1: 102-104.
- Akanji, M.A., O.A. Olagoke and O.B. Oloyede, 1993. Effects of chronic consumption of metal-bisulphate on the integrity of rat liver cellular system. *Toxicology*, 81: 173-179.
- Ayandiran, T.A., O.O. Fawole, S.O. Adewoye and M.A. Ogundiran, 2009. Bio-concentration of metals in the body muscle and gut of *Clarias gariepinus* exposed to sub-lethal concentrations of soap and detergent effluent. *J. Cell Anim. Biol.*, 3: 113-118.
- Chun, W.C. and H.Z. You, 2009. Nonionic surfactant Brij35 effects on toluene biodegradation in a composite bead bio-filter. *Afr. J. Biotechnol.*, 8: 5406-5414.
- DPR, 1991. Environmental Guidelines and standards for the petroleum industry in Nigeria. Ministry of Petroleum Resources, Lagos, UK., pp: 35-76.
- Dehelean, C., V. Nastase, A. Dragomirescu, A. Heges and E. Dinte, 2004. Skin toxicity of sodium lauryl sulfate as evidenced in an animal model. *Rev. Med. Chir. Soc. Med. Nat. Iasi.*, 108: 169-172.
- Easy Essay, 2007. Effects of laundry detergent on the environment. <http://ez-essay.blogspot.com/2007/12/effects-of-laundry-detergent-on.html>.
- Esenowo, I.K. and O.A. Ugwumba 2010. Growth response of catfish (*Clarias gariepinus*) exposed to water soluble fraction of detergent and diesel oil *Environ. Res. J.*, 4: 298-301.
- Ezemonye, L.I.N. and A. Enuneku, 2005. Evaluation of acute toxicity of cadmium and lead to amphibian tadpole (Toad: *Bufo maculatus* and frog: *Ptychadena bibroni*) and frog. *J. Aqua. Sci.*, 20: 33-36.
- Faremi, A.Y. and O.B. Oloyede, 2010. Biochemical and assessment of the effects of soap and detergents industrial effluents on enzymes in the stomach of albino rats. *Res. J. Environ. Toxicol.*, 4: 127-133.
- Gholami, M., S.M.R. Fatemi, M. Falahi, A. Esmaili and A. Mashinchian, 2010. effects of heavy metals (copper and cadmium) and detergent (LAS) on white fish fry *Rutilus frisii* Kutum. *Res. J. Environ. Toxicol.*, 4: 231-236.
- Hasan, F., A.I. Shah, S. Javed and A. Hammed, 2010. Enzymes used in detergent: Lipases. *Afr. J. Biotechnol.*, 9: 4836-4844.
- Kassem, N., 2010. Allergens and toxins in laundry detergents: Synthetic ingredients that are harmful to people and the environment. <http://organizedwisdom.com/allergens-and-toxins-in-laundry-detergents-synthetic-ingredients-that-are-harmful-to-people-and-the-environment/1936071/nxi/med>
- Khurana, R., 2002. Detergents: Counting the cost of cleanliness. *Toxic Link Fact Sheet*, 16: 1-4.
- Lightowers, P., 2004. Still dirty: A review of action against toxic products in Europe. A report of WWF-UK.
- Ogunbileje, J.O. and O.M. Akinosun, 2011. Biochemical and hematological profile in Nigeria cement factory workers. *Res. J. Environ. Toxicol.*,

- Ogundiran, M.A., O.O. Fawole, S.O. Adewoye and T.A. Ayandiran, 2009. Pathologic lesion in the gills of *Clarias gariepinus* exposed to sub-lethal concentrations of soap and detergent. *J. Cell Anim. Biol.*, 3: 78-82.
- Ogundiran, M.A., O.O. Fawole, S.O. Adewoye and T.A. Ayandiran, 2010. Toxicological impacts of detergent effluent on juvenile of African Catfish (*Claris gariepinus* B.). *Agric. Bio. Am.*, 1: 330-342.
- Ojo, O.A. and B.A. Oso, 2009. Biodegradation of synthetic detergents in wastewater. *Afr. J. Biotechnol.*, 8: 1090-1109.
- Redmond, W.A., 2011. Detergent: Encarta microsoft corporation (DVD). USA.
- Rejeki, S., D. Desrina and R.A. Mulyana, 2006. Chronic affects of detergent surfactant (Linear alkylbenzene sulfonate/LAS) on the growth and survival rate of sea bass (*Lates calcalifer* Bloch) larvae. Aquaculture Study Program Fisheries Department, Faculty of Fisheries and Marine Science, Diponegoro University, Semarang, Indonesia, pp: 1-18. http://eprints.undip.ac.id/91/1/chronic_affects_of_detergent_surfactant.pdf.
- Riaz, S., A. Ahmad and S. Hasnain, 2009. Antibacteria activity of soaps against daily encountered bacteria. *Afr. J. Biotechnol.*, 8: 1431-1436.
- Ronni, W., 2004. Detergent Chemistry: Foundations and Application: Encyclopedia. Com. Accessed 15 May, 2011 at <http://www.encyclopedia.com>.
- Taylor, D.J., N.P.U. Green and G.W. Stout, 2003. Biological Science. 3rd Edn., Cambridge University Press, UK., pp: 378.
- Uaboi-Egbenni, P.O., P.N. Okolie, O.E. Adejuyitan, A.O. Sobande and O. Akinyemi, 2009. Effects of industrial effluents on the growth and anatomical structures of *Abelmoschus esculentus* (Okro). *Afr. J. Biotechnol.*, 8: 3251-3260.
- Wadaan, M.A.M. and M. Mubarak, 2009. Blood chemistry changes as an evidence of the toxic effects of anionic surfactant sodium dodecyl sulfate. *Asian J. Scientific Res.*, 2: 113-118.
- Warne, M.J. St. and A.D. Schiffko, 1999. Toxicity of laundry detergent components to a freshwater cladoceran and their contribution to detergent toxicity. *Ecotoxicol. Environ. Safety*, 44: 196-206.