



Biolog. Journal of Armenia, 1-2 (72), 2020

EFFECT OF OIL PRODUCING ACTIVITIES ON BIOCHEMICAL INDICES OF NIGERIAN DWARF GOAT

FIDELIS IFEAKACHUKU ACHUBA

Department of Biochemistry, Delta State University, PMB 1, Abraka Nigeria
achubbcha@yahoo.com, achuba@delsu.edu.ng

The implication of crude oil producing activities on animals and humans in the Niger-Delta attracts the attention of various scientists. The aim of this investigation was to access the biochemical indices of native goat inhabiting crude oil producing environment using nondestructive sampling techniques. Similar aged matched goats from a non-oil producing environment were adopted as control. Blood samples from each set of goats were collected using hypodermal needle through the ear vein of each goat. The sera were prepared and used to determine NADPH oxidase activity, lipid peroxidation product (MDA), reduced glutathione (GSH) level, ascorbic acid (AA) level, and glucose-6-phosphate dehydrogenase (G6PD) activity. The results indicated that the activity of NADH oxidase activity and lipid peroxidation products were higher in goat obtained from the oil producing environment compared to values in goat from non-oil producing areas. This is against the reduced levels of antioxidant markers: GSH, AA and G6PD in goats inhabiting oil producing area relative to those in the non-oil producing region. This study thus concluded that animals living in area close to crude oil producing activities are faced with pollution stimulated health challenges.

*Ascorbic acid – crude oil – glutathione – lactate dehydrogenase –
Glucose-6-phosphate dehydrogenase*

Նիգեր-դելտայում կենդանիների և մարդկանց վրա հում նավթի արդյունահանման գործողությունների հետևանքները գրավում են տարբեր գիտնականների ուշադրությունը: Այս հետաքննության նպատակն էր մուտք գործել հում նավթ արտադրող միջավայրում բնակվող հայրենի այծի կենսաքիմիական ցուցանիշներ՝ օգտագործելով ոչ քանդման նմուշառման մեթոդներ: Յուր չարտադրող միջավայրից նման տարիքի համապատասխանեցված այծերն ընդունվել են որպես հսկողություն: Յուրաքանչյուր այծի արյան նմուշները հավաքվում էին հիպոդերմիկ ասեղով՝ յուրաքանչյուր այծի ականջի միջոցով: Սերաները պատրաստվել և օգտագործվել են NADPH-ի օքսիդազի ակտիվությունը, լիպիդային պերօքսիդացման արտադրանքը (MDA), գլյուտաթիոն (GSH) մակարդակի իջեցումը, ասկորբինաթթվի (AA) մակարդակը և գլյուկոզա-6-ֆոսֆատ դեհիդրոգենազ (G6PD) գործունեությունը որոշելու համար: Արդյունքները ցույց են տվել, որ NADH-ի օքսիդազի գործունեության ակտիվությունը և լիպիդային պերօքսիդացման արտադրանքի ակտիվությունն ավելի բարձր են յուր արտադրող միջավայրից ստացված այծի մեջ՝ համեմատած յուր չարտադրող տարածքներից այծի արժեքների հետ: Սա դեմ է հակաօքսիդանտային մարկերների իջեցված մակարդակի. GSH, AA և G6PD՝ նավթարդյունահանող տարածք բնակեցրած այծերում, համեմատած նավթ չարտադրող տարածաշրջանում: Այս ուսումնասիրությունը եզրակացրեց, որ անմշակ յուր արտադրող գործունեությանը հարող տարածքում ապրող կենդանիները բախվում են աղտոտման խթանման առողջության հետ կապված մարտահրավերներին:

*Ասկորբինաթթու – չմշակված յուր – գլյուտաթիոն – կաթնաթթվային ջրազերծում –
գլյուկոզա-6-ֆոսֆատ դեհիդրոգենազ*

Влияние деятельности по добыче сырой нефти на животных и людей в дельте реки Нигер привлекает внимание различных ученых. Цель этого исследования состояла в том, чтобы получить доступ к биохимическим показателям местных коз, обитающих в нефтесодержащей среде, с использованием методов неразрушающего отбора проб. Подобные состарившиеся козы из нефтесодержащей среды были приняты в качестве контроля. Образцы крови из каждой группы коз отбирали с помощью иглы для подкожных инъекций через ушную вену каждой козы. Сыворотки готовили и использовали для определения активности NADPH-оксидазы, продукта перекисного окисления липидов (MDA), уровня глутатиона (GSH), уровня аскорбиновой кислоты (AA) и активности глюкозо-6-фосфатдегидрогеназы (G6PD). Результаты показали, что активность NADH-оксидазы и продуктов перекисного окисления липидов была выше у коз из нефтесодержащей среды, по сравнению со значениями у коз из нефтесодержащих районов. Это противоречит снижению уровня антиоксидантных маркеров: GSH, AA и G6PD у коз, населяющих нефтесодержащий район, по сравнению с районами, не производящими нефть. Таким образом, это исследование пришло к выводу, что животные, живущие в районе, близком к добыче сырой нефти, сталкиваются с проблемами, вызванными загрязнением.

Аскорбиновая кислота – сырая нефть – глутатион – лактатдегидрогеназа – глюкозо-6-фосфатдегидрогеназа

During the exploration and production of crude oil, hydrocarbons of various sorts, carbon (IV) oxide, corrosive acid wastes, sulphur and toxic metals are released into the immediate surrounding [39]. The biological consequences of this have been monitored by field and laboratory studies [1, 22]. Some of the noxiousness on plants include induction of oxidative stress [1, 17]; alteration of growth and metabolic activities in plants [10, 13, 15, 16, 20]; induction of metabolic derangements in animals [1, 7, 9, 12, 16, 17].

Most importantly, the health implication of crude oil activities in the Niger Delta is enormous [26-28]. Also, environmentally mediated alterations in biochemical indices have been reported [9, 34]. Overall, crude oil pollution is a major threat to all shades of lives in the Niger Delta Region. That if adequate care is not taken may culminate in the extinction of some plants and animal species. Recently, the treat of industrial activities on biodiversity in Delta State was documented [25]. The focus of this investigation was to assess the effect of crude oil related activities on Nigerian dwarf goat.

Materials and methods. Experimental Animals and collection of Samples

Mature female goats, twelve from each location were used for this study. Nigerian goat inhabiting two regions: Okpai and its environs with a known history of oil activities in Ndokwa East Local Government Area, Delta State, Nigeria and Eziokpor and its environs in Ukwuani Local Government also in Delta State, Nigeria with no history of crude oil activities were chosen for the study. After due consent of the owners were sought, sterile hypodermal syringe were used to collect blood samples through the ear vein into sample containers and labeled appropriately. The samples were kept in ice and taken to the laboratory where they were stored at 4°C and analyzed within forty eight hours.

Determination of Biochemical parameters

Red blood cells were isolated as described by George et al. [25] and used for NADPH oxidase was determined following the protocol of Jiang and Zhang [35]. Thiobarbituric acid-reactive substances (TBARS) formation was measured as index of lipid peroxidation [27].

Vitamin C was determined as reported by Achuba [18]. Serum reduced glutathione concentration was determined with the method of Ellman [33]. Blood GSSG was determined with the method described by Tietze [48]. The ratio of GSH/GSSG was evaluated with the equation = $GSH - 2GSSG / GSSG$. Glucose-6-phosphate dehydrogenase activity was determined by the method of Henry [33].

Results and Discussion. The ingestion of diet polluted by petroleum causes the release of reactive intermediates which induce changes in tissues of the organism in question [4, 43]. However, exposed organisms tend to adjust metabolic indices in a bid to cope with the effects of pollution. That the animals inhabiting the studied area are experiencing free radical toxicity is indicated by the upsurge in the activity of NADPH oxidase in areas associated with petroleum pollution (fig.1).

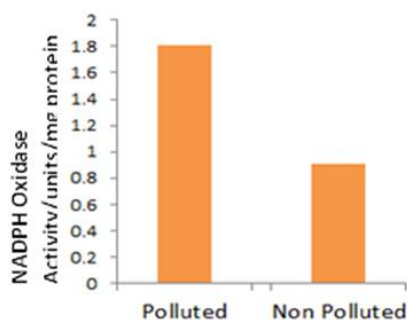


Fig. 1. NADPH oxidase activities in animals from polluted and non-polluted environments. The mean serum NADPH oxidase activity of the animals differs significantly ($p < 0.05$) in relation to environments

This enzyme is a potent free radical generator [46]. Free radicals when produced in excess than the organism can contain results in biomembrane damages occasioned by radical mediated macromolecular damage. One such mediator in membrane damage is lipid peroxidation. Environment-mediated lipid peroxidation alterations had been reported [8]. It makes no surprise the increase in lipid peroxidation products in animals in the perceived polluted environment (fig. 2).

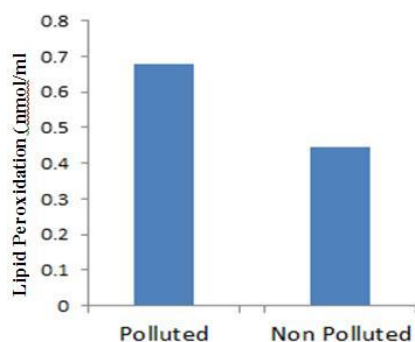


Fig. 2. Lipid peroxides products in animals from polluted and non-polluted environment. The mean serum lipid peroxides of the animals differs significantly ($p < 0.05$) in relation to environments

Lipid peroxidation has a very strong relationship with induction of disease cum enzyme modulations [1, 11]. One important enzyme that gives information on respiratory hiccups in aerobic organisms is lactate dehydrogenase (LDH). The activity of the enzyme increases when there is a shift towards anaerobiosis [23]. Therefore, the increase in LDH of animals from polluted environment is a reflection pollution-stimulated anaerobic respiration (fig. 3).

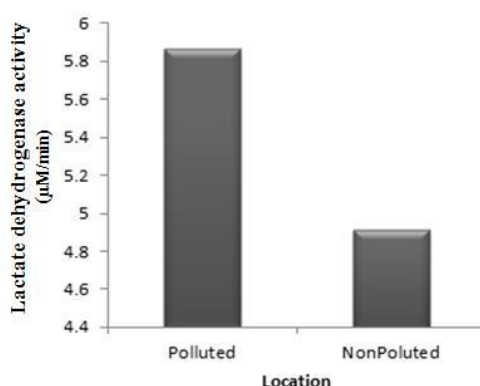


Fig. 3. Lactate dehydrogenase activities (µM/min) in animals from polluted and non-polluted environment. The mean serum lipid peroxides of the animals differs significantly ($p < 0.05$) in relation to environments

It is no gainsaying; therefore, that animals exposed to polluted environments are prone to infections since earlier report implicated LDH as a mediator in disease prognosis [47]. An array of non-enzymatic antioxidants such as vitamin C and glutathione are altered during oxidative insults [18, 44]. Ascorbic acid is a water-soluble antioxidant vitamin, which scavenges free radicals in the cytosol by donating electrons to free radicals to inactivate them [45]. This study indicated a reduction in serum ascorbic acid concentration in animals as a result of the polluted environment (fig. 4).

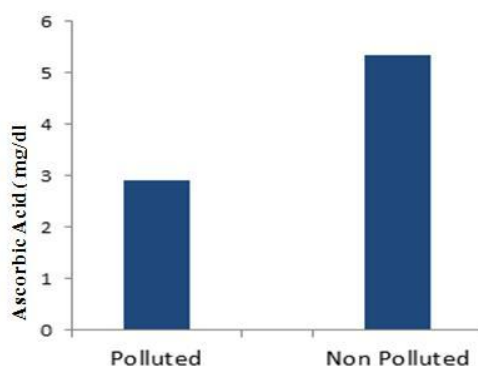


Fig.4. Level of ascorbic acid in the serum of animals from polluted and non-polluted environment. The mean serum ascorbic acid level of the animals differs significantly ($p < 0.05$) in relation to environments.

This observation is similar to previous report by Achuba [18]. This significant reduction in AA is no surprise putting into consideration the increase in serum lipid peroxidation product (fig. 3). It is pertinent to posit that the low level of AA in animals from the polluted areas is simply due to the utilization of ascorbic acid in scavenging the reactive intermediates generated in the tissue of animals exposed to pollution. Further depletion of the non-enzymatic antioxidant systems is also indicated by the depletion of GSH in animals obtained from the polluted area (fig. 5).

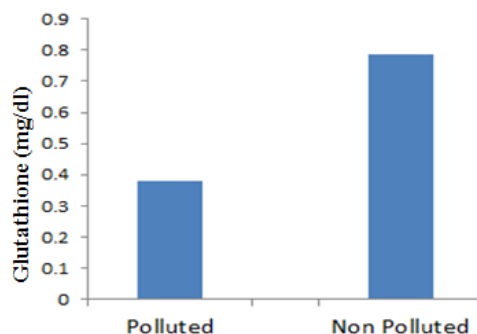


Fig.5. Level of reduced glutathione in the serum of animals from polluted and non-polluted environment. The mean serum reduced glutathione level of the animals differs significantly ($p < 0.05$) in relation to environments

This reduction could predispose the animals from polluted areas to oxidative damage. Similar in action but different in distribution, glutathione is abundant in the mitochondria and is the major soluble antioxidant in this cellular compartment [37]. In fact, glutathione level was affected by the pollution in the environment. Recently, Adeoye et al. [24] that pollution depletes the level of glutathione in inhabiting animals. Similar to AA, an upsurge in reactive intermediates in tissues of the animals due pollution might the basis for the displayed reduction in GSH concentrations and the concurrent decrease in GSH/ GSSG ratio in animals inhabiting the polluted environment (fig. 6).

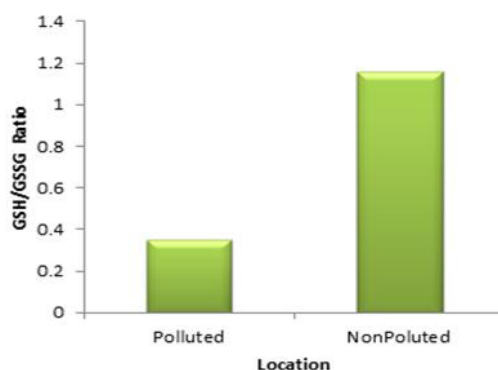


Fig. 6. GSH/ GSSG ratio in animals from polluted and non-polluted environment. The mean serum ratio of GSH/GSSG of the animals differs significantly ($P < 0.05$) in relation to environments.

The ratio of reduced glutathione (GSH)/ oxidized glutathione (GSSG) is a good candidate for measuring oxidative stress in living organisms. And increase in the ratio of GSSG/GSH portends oxidative stress [36, 38]. The decrease in the ratio of GSH/GSSG in animals in the polluted relative to animals in the non-polluted environment is indicative of pollution-stimulated oxidative stress.

Another important enzyme related to glutathione is G6PD whose activity was low in animals from the polluted sites relative to animals taken from the non-polluted regions (fig. 7).

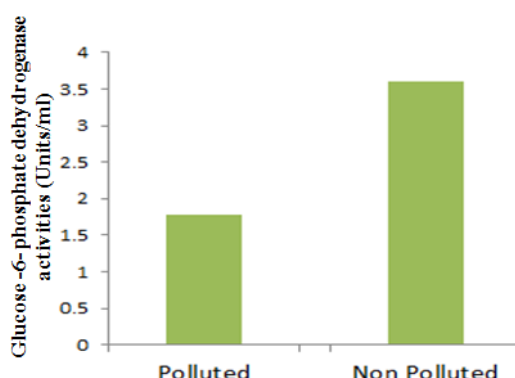


Fig. 7. Glucose -6- phosphate dehydrogenase activities in animals from polluted and non-polluted environment

G6PD is the first important enzyme in the pentose phosphate pathway that produces NADPH that is required for maintaining glutathione in its reduced state [32]. This may explain the crucial role of this enzyme in the prevention of oxidative damage [29, 40]. The reduced activity of this enzyme in animals from polluted environment further confirm the reason for reduced glutathione and the enhanced GSH/GSSG in animals in the polluted environment relative to animals in the non-polluted environment

The exposure to chronic petroleum pollution has an adverse biochemical consequence in animals inhabiting polluted environment. This is evidenced by the negative alterations of oxidative stress indices in animals used in this investigation.

REFERENCES

1. Achuba F.I. Role of bitter leaf (*Vernonia amygdalina*) extract in prevention of renal toxicity induced by crude petroleum contaminated diets in rats. *International journal of veterinary science and medicine*, 6, 2, 172-177, 2018a .
2. Achuba F.I. Protective Influence of *Elaeagnus guineensis* leaf in diet on petroleum-mediated kidney damage in rat. *Nigerian Journal of Pharmaceutical and Applied Science Research*, 7, 2, 33-38, 2018b.
3. Achuba F.I. Powdered Oil Palm (*Elaeagnus guineensis* Jacq) leaf as remedy for hydrocarbon induced liver damage in Rats. *Nigerian Journal of Pharmaceutical and Applied Science Research*, 7, 3, 89-95, 2018c.
4. Achuba F.I. Bitter leaf (*Vernonia amygdalina* Del) extract potentiates testicular metabolic stress induced by petroleum-tainted diets in rats. *Nigerian Journal of Pharmaceutical and Applied Science Research*, 8, 1, 44-51, 2018d.
5. Achuba F.I. Modulation of crude oil induced alteration of oxidative stress indices in rat by red palm oil. *Journal of Applied Sciences and Environmental Management*, 22, 6, 929-932, 2018e.
6. Achuba F.I., Ebokaiwe P., Peretiemo-Clarke B.O. Effect of Environmental Pollution on Oxidative Stress in African Catfish (*Clarias heterobranchus*), *International Journal of Environmental Monitoring and Analysis*. 2, 6, 297-301. doi:10.11648/j.ijema.20140206.11 2014.
7. Achuba F.I., Awhin P.E. Protective influence of antioxidant vitamins on hematological indices of rabbits fed crude-oil-contaminated diet. *Toxicological and Environmental Chemistry*, 9, 3, 505-510, 2009.

8. *Achuba F.I.* Superoxide dismutase and Lipid peroxidation levels in fish from the Ethiopie River in Southern Nigeria. *Bulletin of Environmental Contamination and Toxicology*, 69, 6, 892-899, 2002.
9. *Achuba F.I., Ogwumu M.D.* Possible protective role of palm oil and beef liver on the kidney and liver of wistar albino rats fed diesel-contaminated diet. *Biokemistri*, 26, 4, 129, 2014.
10. *Achuba F.I.* The effects of sublethal concentrations of crude oil on the growth and metabolism of cowpea (*Vigna unguiculata*) seedlings. *The Environmentalist*, 26, 1, 17-20, 2006.
11. *Achuba F.I., Osakwe S.A.* Petroleum induced free radical toxicity in Africa catfish (*clarias gariepinus*) *Fish Physiology and Biochemistry*, 29, 2, 97-103, 2003.
12. *Achuba F.I. Peretiomo – Clarke B.O, Okolie T.C.* Oxidative stress in the brain of rabbits with petroleum- induced hypoglycaemia. *Biological letters*, 42, 1, 33-39, 2005.
13. *Achuba F.I., Ohwofasa S.O.* Influence of fish pond wastewater treatment of diesel tainted soil on metabolic activities of cowpea (*Vigna unguiculata*) seedlings. *FUW Trends in Science Technology Journal.*, 4, 1, 169-173, 2019.
14. *Achuba F. I.* Petroleum products in soil mediated oxidative stress in cowpea (*Vigna unguiculata*) and maize (*Zea mays*) seedlings. *Open Journal of Soil Science*, 4, 12, 417-435, 2015.
15. *Achuba F.I., Ja-anni M.O.* Effect of abattoir waste water on metabolic and antioxidant profiles of cowpea seedlings grown in crude oil contaminated soil. *Int. J. Recycl. Organic Waste Agric*, 7, 1, 59 -66, 2018.
16. *Achuba F.I.* Effect of vitamins C and E intake on blood lipid concentration, lipid peroxidation, superoxide dismutase and catalase activities in Rabbit fed petroleum contaminated diet. *Pakistan Journal of Nutrition*, 4, 5, 330-335, 2005.
17. *Achuba F. I., Nwokogba C.C* Effects of honey supplementation on hydrocarbon-induced kidney and liver damage in wistar albino rats. *Biokemistri*, 27, 1, 50-55, 2015.
18. *Achuba F. I.* African land snail *Achatina marginatus*, as bioindicator of environmental pollution. *North- Western Journal of Zoology*, 4, 1, 1-5, 2008.
19. *Achuba F.I. Iserhienrhien L.O.* Effects of soil treatment with abattoir effluent on morphological and biochemical profiles of cowpea seedlings (*V. unguiculata*) grown in gasoline polluted soil. *Ife Journal of Science*, 19, 3, 051-059, 2018.
<https://dx.doi.org/10.4314/ijfs.v20i1.5>
20. *Achuba F.I., Erhijivwo P.O.* The effect of abattoir waste water on the metabolism of cowpea seedlings grown in diesel contaminated soil. *Nigerian Journal of science and Environment*, 15, 1, 155-162, 2017.
21. *Achuba F.I., Peretiomo-Clarke B.O.* Effect of spent engine oil on soil catalase and dehydrogenase activities. *International Agrophysics*, 22, 1, 1-4, 2008.
22. *Achuba F.I., Ekute B.O.* Effect of exposure to chronic petroleum pollution on biomarkers of oxidative stress in African toad (*Bufo regularis*) in Parts of Delta State, Nigeria *International Journal of Research in Pharmacy and Biosciences*, 6, 7, 18-236, 2019.
23. *Achuba F.I.* Effect of petroleum products treatment of soil on succinate dehydrogenase and lactate dehydrogenase activities in cowpea (*Vigna unguiculata*) and Maize (*Zea mays*) seedlings *American Journal of Experimental Agriculture*, 5, 5, 498, 2015b.
24. *Adeoye O., Olawumi, J., Opeyemi, A., Christiania O.* Review on the role of glutathione on oxidative stress and infertility. *JBRA assisted reproduction*, 22, 1, 61-66, 2018.
[doi:10.5935/1518-0557.20180003](https://doi.org/10.5935/1518-0557.20180003)
25. *Ajani F., Pudie A.* Review of Industrial Activities: A Threat to Biodiversity and Ecotourism Development in Delta State, Nigeria, 6, 1, 8-20, 2019 . [doi:10.30845/jals.v6n1p2](https://doi.org/10.30845/jals.v6n1p2)
26. *Atubi A.O., Onu M.A.* The Effects of Carbon Black on Human Health in Ubeji, Shaguolo and Environs, Delta State, Nigeria, *International Journal of Medical Science and Clinical invention* 5, 11, 4162-4177, 2018.
<https://doi.org/10.18535/ijmsci/v5i11.05>
27. *Atubi A.O.* Effects of oil spillage on human health in producing communities of Delta State, Nigeria. *European Journal of Business and Social Sciences*, 4, 8, 14-30, 2015.
28. *Bruederle A., Hodler R.* The Effect of Oil Spills on Infant Mortality: Evidence from Nigeria, *CESifo Working Paper*, No. 6653, 2017.

29. Cramer C.T., S. Cookie, L.C. Ginsberg, R.F. Kletzien, S.R. Stapleton, R.G. Ulrich, Upregulation of glucose-6-phosphate dehydrogenase in response to hepatocellular oxidative stress: Studies with diquat. *J. Biochem. Toxicol.*, 10, 6, 293-298, 1995.
30. Ellman G.C. Tissue sulfhydryl groups. *Arch. Biochem. Biophys.*, 82, 70-77, 1959.
31. George A., Pushkaran S., Konstantinidis D. G., Koochaki S., Malik P., Mohandas N., Kalfa T.A. Erythrocyte NADPH oxidase activity modulated by Rac GTPases, PKC, and plasma cytokines contributes to oxidative stress in sickle cell disease. *Blood*, 121, 11, 2099-2107, 2013. doi:10.1182/blood-2012-07-441188.
32. Halliwell B., J.M.C. Gutteridge, Free Radicals in Biology and Medicine. 4th Edn., Oxford University Press, Oxford, p.p. 851, 1998.
33. Henry J.B. Clinical Diagnosis and Management by Laboratory Methods W.B. Saunders and Company, Philadelphia, PA p. 365, 1979.
34. Isamah G.K., Asagba S.O. A Comparative study on the activities of xanthine oxidase and aldehyde oxidase in different fish species from two rivers in the western Niger-Delta. *Environ Monit Assess* (2004) 91, 1-3, 293-300, 2004. <https://doi.org/10.1023/B:EMAS.0000009242.94035.a4>
35. Jiang M., Zhang J. Involvement of plasma-membrane NADPH oxidase in abscisic acid and water stress-induced antioxidant defense in leaves of maize seedlings. *Planta*, 215, 6, 1022-2002.
36. Knight T.R., Kurtz, A. Bajt, M.C., Hinson, J.A. and Jaeschke, H., Vascular and Hepatocellular peroxynitrite formation during acetaminophen induced liver injury, Role of mitochondrial oxidant stress. *Toxicology Science*. 62, 212-220, 2001.
37. Masella R., D: Benedetto R, Yan R., Filesi C., Giovannini C. Novel mechanisms of natural antioxidant compounds in Biological systems: Involvement of glutathione related enzymes. *J. Nutri. Biochem.*, 16, 577-586, 2005.
38. Németh I., Boda D. The ratio of oxidized/reduced glutathione as an index of oxidative stress in various experimental models of shock syndrome. *Biomed Biochim Acta*. 48, 2-3, S53-7, 1989.
39. Ogeleka D.F., Tudararo-Aherobo L.E., Okieimen F.E. Ecological effects of oil spill on water and sediment from two riverine communities in Warri, Nigeria. *Int. J. Biol. Chem. Sci.*, 11, 1, 453-461, 2017.
40. Oh S.I., Kim C.I., Chun H.J., Park, S.C, Chronic ethanol consumption affects glutathione status in rats. *Journal of Nutrition*, 128, 4, 758-763, 1998.
41. Ohkawa H., Ohishi N., Yagi K. Assay for lipid peroxides in animal tissues by Thiobarbituric acid reaction. *Anal of Biochemistry*, 95, 2, 351-8, 1979.
42. Okoro I.O. Role of antioxidants in the palatability and production potentials of water yam (*Dioscorea alata* L. cv kurudu) *Electronic Journal of Environmental, Agricultural and Food Chemistry*, 9, 2, 364-368, 2010.
43. Okpoghono J, George B.O., Achuba F.I. Assessment of antioxidant indices after incorporating crude oil contaminated Catfish and African nutmeg (*Monodora myristica*) extracts into rat diet. *Journal of Applied Sciences and Environmental Management*, 22, 2, 197-202, 2018.
44. Okpoghono J, Achuba F.I., George B.O. Protective effects of *Monodora myristica* extracts on crude petroleum oil-contaminated catfish (*Clarius gariepinus*) diet in rats. *International Journal of Veterinary Science and Medicine*, 6, 117-122, 2018a.
45. Pehlivan F.E. Vitamin C: An Antioxidant Agent, Vitamin C, Amal H. Hamza, IntechOpen, D, 2017. OI: 10.5772/intechopen.69660. Available from: <https://www.intechopen.com/books/vitamin-C/vitamin-C-an-antioxidant-agent>
46. Segal A.W. The function of the NADPH oxidase of phagocytes and its relationship to other NOXs in plants, invertebrates, and mammals. *The International Journal of Biochemistry & Cell Biology*, 40, 4, 604-618. doi:10.1016/j.biocel.2007.10.003, 2008.
47. Teke H.U., Başak M, Teke D., Kanbay M. Serum Level of Lactate Dehydrogenase is a Useful Clinical Marker to Monitor Progressive Multiple Myeloma Diseases: A Case Report Turk. J. Haematol., 2014 Mar; 31, 1, 84-87, 2014. doi: 10.4274/Tjh.2013.0044.
48. Tietz N.W. Fundamentals of clinical chemistry, W B Saunders Co. Philadelphia. page 723, 1976.

Received on 18.09.2019