

Original Article

Assessment of the Protective Effects of Vitamin C and E on Cypermethrin-induced Nephrotoxicity and Electrolyte Imbalance in Wistar Rats

Johnson Olaleye Oladele^{1,3}, Omowumi Oyeronke Adewale², Olu Israel Oyewole^{1*}, Abiola Gbolagbade¹, Moses Oyedotun Oyeleke^{1,3}

¹Phytomedicine and Toxicology Laboratories, Department of Biochemistry, Faculty of Basic and Applied Sciences, Osun State University, Osogbo, Nigeria.

²Cancer Research and Molecular Toxicology Laboratories, Department of Biochemistry, Faculty of Basic and Applied Sciences, Osun State University, Osogbo, Nigeria

³Biochemistry Unit, Department of Chemical Sciences, Faculty of Sciences, Kings University, Ode-Omu, Nigeria.

*Corresponding Author: ioluoye@yahoo.com

Received: 27-9-2019
Revised: 3-1-2020
Published: 22-1-2020

Keywords:

*Nephrotoxicity,
Cypermethrin,
Vitamins,
Electrolyte Imbalance,
Insecticide*

Abstract: Cypermethrin is a potent pyrethroids insecticide causing different pathological features when exposed to mammal. Vitamins are used as nutrient supplements and in clinical studies as medical intervention in some disease conditions. This study was designed to investigate the possible protective effect of vitamin C and E on Cypermethrin induced nephrotoxicity in wistar albino rats. Twenty-eight (28) wistar albino rats were sorted into four groups of seven rats per groups were used in this study. Group A serves as the control and received distilled water orally. Group B, C and D were administered 25mg/kg body weight cypermethrin orally. Group C and D were treated daily with 40mg/kg body weight vitamin C and 20mg/kg body weight vitamin E respectively by oral administration while group B was left untreated for 14 days. Cypermethrin significantly ($P<0.05$) induced nephrotoxicity as characterized with significant increased ($P<0.05$) in the serum levels of Urea, uric acid and creatinine. It also caused significant decrease ($P<0.05$) in renal total protein, albumin and globulin. Exposure to Cypermethrin induced electrolyte imbalance in rats with significant increase in serum chloride ion, potassium ion and significant decrease in serum level of sodium ion and bicarbonates. Histological results revealed that cypermethrin caused distortion in histoarchitecture of the kidney characterized by lesion of glomerulus, damaged Bowman's capsule, degenerated and vacuolated renal tubules. Taken together, vitamin C and E significantly reverse all these alterations and offer protection to the kidney membrane.

Cite this article as: Oladele, J.O., Adewale, O.O., Oyewole, O.I., Gbolagbade, A., Oyeleke, M.O. (2020) Assessment of the Protective Effects of Vitamin C and E on Cypermethrin-induced Nephrotoxicity and Electrolyte Imbalance in Wistar Rats. *Journal of basic and applied Research in Biomedicine*, 6(1): 1-6



This work is licensed under a Creative Commons Attribution 4.0 License. You are free to copy, distribute and perform the work. You must attribute the work in the manner specified by the author or licensor.

INTRODUCTION

Pyrethroids are synthetic analogues of pyrethrins, the active substances in the flowers of *Chrysanthemum*. Pyrethroids can be classified into two large groups. Type I pyrethroids which do not contain a cyano group in their molecules such as tetramethrin, permethrin, and phenothrin. Type II pyrethroids contain a cyano group such as deltamethrin, cyphenothrin, cypermethrin, and fenvalerate. Mammalian toxicity of the two types of pyrethroids causes different pathological features. Exposure to type I pyrethroids induced hyper excitation, ataxia, convulsions, and eventual paralysis while type II pyrethroids toxicity is characterized by hypersensitivity, choreoathetosis, tremors, and paralysis.

Cypermethrin is a potent pyrethroids insecticide and first synthesized by Elliott et al., (1974). It is a highly active synthetic insecticide used against a wide range of pests in agriculture, public health and animal husbandry to increase the production of food grains and other agricultural-products (Usmani and Knowles, 2001). There is increased

risk of food being contaminated with the insecticide, which may harm humans and domesticated animals. Cypermethrin produces drastic effects on both the invertebrates (Gowland et al., 2002) and vertebrates (Das and Mukherjee, 2003).

The prophylactic use of vitamins as medical intervention in some pathological conditions could be due to their ability to mitigate against oxidative stress mediated by these diseases. Vitamins such as vitamin C and E have many biological functions, the antioxidant function being the most important and best known (Bell, 1987). They are used as nutrient supplementation in clinical studies and are being widely used by people with various kinds of diseases in order to partially treat, retard or slow down the deterioration effects of these disorders. Other functions of these vitamins include enhancement of enzymatic activities, gene expression, and neurological function(s). The most important function of vitamin E has been suggested to be in cell signalling. As an antioxidant, vitamin E acts as a peroxy radical scavenger, preventing

the propagation of free radicals in tissues, by reacting with them to form a tocopheryl radical, which will then be reduced by a hydrogen donor (such as vitamin C) and thus return to its reduced state (Traber and Stevens, 2011).

This study is therefore designed to investigate the benefit effects of vitamin C and E against Cypermethrin mediated nephrotoxicity in female wistar rats.

MATERIALS AND METHODS

Chemicals/Reagents

Cypermethrin [Cyano-(3-phenoxyphenylmethyl) 3-(2,2-dimethylpropane-1-carboxylate)] is obtained as Cyforce, a product of Gharda Chemicals limited India, Vitamin C is obtained from Jinling pharmaceuticals, Vitamin E is a product of Embassy pharmaceuticals, albumin, total protein, bilirubin, urea, creatinine assay kits are products of Randox Chemical limited, England. All other chemicals are of analytical grade and were obtained from Analar BDH Limited, Poole, England.

Experimental Protocol

This study was performed with twenty-eight female albino rats (average weights 150-160g) obtained from the Department of Anatomy, University of Ibadan and were housed in ventilated cages in the Animal house of Biochemistry Department, Osun State University, Osogbo, Nigeria. The rats were acclimatized for 3 weeks before administration of the drugs. The experiment was carried out in accordance to current rules and guidelines that have been established for the care of the laboratory animals (NRC, 2011).

The animals were divided into four groups of seven rats per groups. Group A serves as the control and received distilled water orally. Group B, C and D were administered 25mg/kg body weight cypermethrin orally. Group C and D were treated daily with 40mg/kg body weight vitamin C and 20mg/kg body weight vitamin E respectively by oral administration while group B was left untreated. Animals were kept at optimum temperature with a 12 hours light/dark cycle and given rat feed in form of pellet and water ad libitum. The cages were cleaned twice daily. The period of administration lasted for 14 days.

Preparation of serum

The rats were sacrificed 24 hrs after the last treatment by cervical dislocation. The jugular vein was cut and blood sample collected into clean, dry centrifuge tube. The blood was left for 10 min at room temperature to clot after which it was centrifuged at 4,000 rpm in an MSC (Essex, UK) bench centrifuge. The clear supernatant (serum) was aspirated using a Pasteur pipette into clean, dry

sample bottles and then stored at -4°C for biochemical analyses.

Preparation of tissue homogenates

The kidney was quickly excised from the rat and immediately placed on a blotting paper to remove blood stains. The tissues were then rinsed in 1.15% KCl to remove haemoglobin followed by homogenization in 4 volumes of ice-cold 0.01 M potassium phosphate buffer (pH 7.4) using Teflon homogenizer. The homogenates were centrifuged at 12,500 g for 20 min at 4 °C to obtain supernatants (post-mitochondrial fractions) which were stored till required for assay.

Determination of biochemical parameters

Serum creatinine, urea and uric acid were determined colorimetrically as earlier described by Cheesbrough (2005). Colorimetric determination of protein concentration was done using bovine serum albumin as standard (Lowry et al., 1951) while serum globulin was estimated using the method of Mokady et al. (1989). Albumin concentration in the serum was measured using the bromocresol green method (Tietz et al., 1994).

Histological Examination

The kidneys were fixed in 10% formalin and embedded in paraffin wax. Thin sections (7–9 mm thickness) of the tissues were cut and dewaxed in xylene, hydrated in decreasing percentage of alcohol and stained with hematoxylin. They were then dehydrated in increasing percentage of alcohols till 70% and stained with 1% alcoholic eosin. They were differentiated in 90% alcohol and cleared in xylene. These stained sections were observed under the microscope for histopathological analysis.

Statistical analysis

Results obtained were expressed as mean value \pm standard deviation (SD). Comparison was done using one-way analysis of variance (ANOVA) between the control and treatment groups. P values <0.05 were considered statistically significant.

RESULTS

The effect of administration of vitamin C and E on cypermethrin -induced changes in serum urea, creatinine and uric acid concentrations in female wistar rats

Cypermethrin induced significant alterations in serum urea, creatinine and uric acid concentrations in the exposed rats when compared to control, administration of vitamins C and E individually significantly altered these results (fig 1).

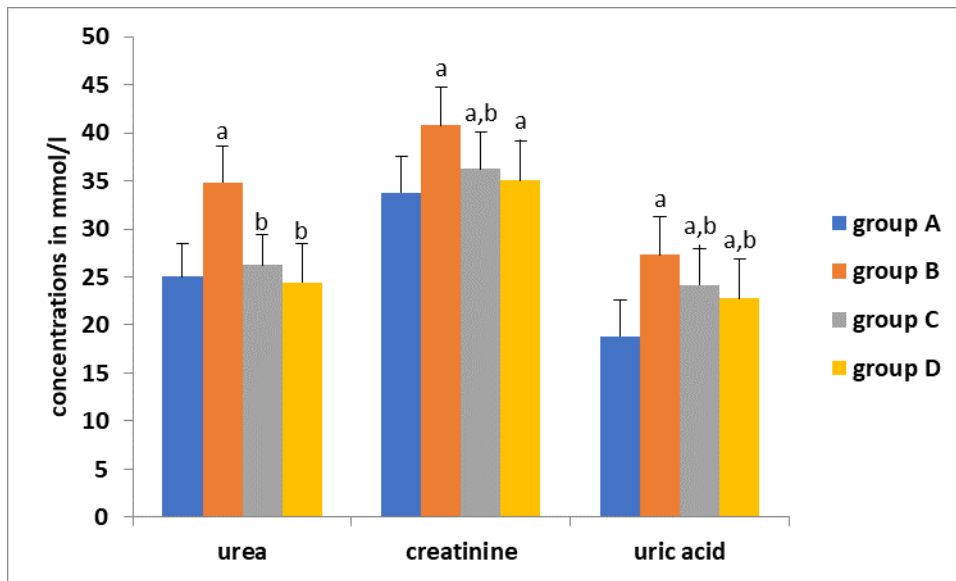


Fig 1. The effect of administration of vitamin C and E on cypermethrin -induced changes in serum urea, creatinine and uric acid in female wistar rats. Values are means \pm SD. ^a values are significantly different from normal control (group A) at $P < 0.05$. while ^b values are Significantly different from cypermethrin group (group B) at $P < 0.05$.

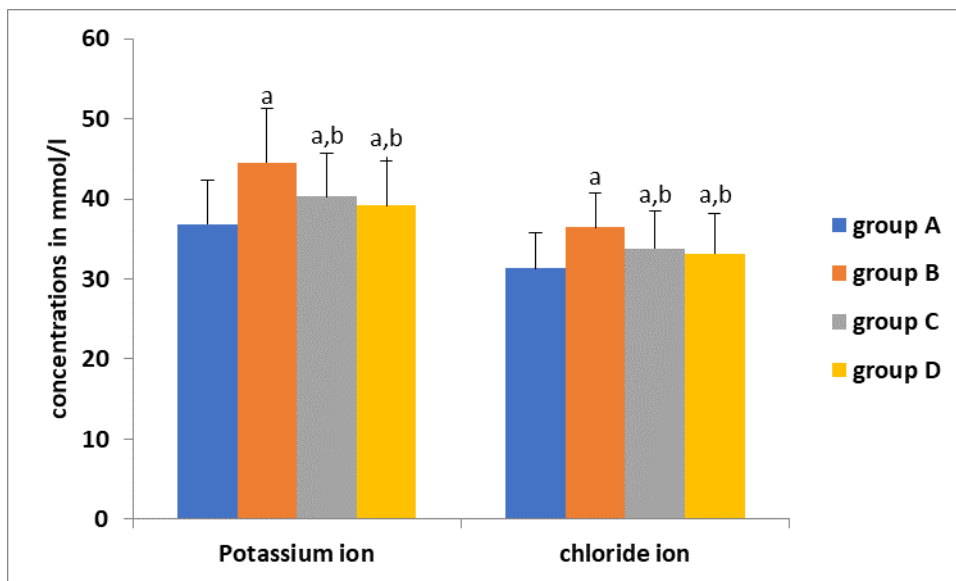


Fig 2. The effect of administration of vitamin C and E on cypermethrin -induced changes in serum potassium ion and chloride ion concentrations in female wistar rats. Values are means \pm SD. ^a values are significantly different from normal control (group A) at $P < 0.05$. while ^b values are Significantly different from cypermethrin group (group B) at $P < 0.05$.

The effect of administration of vitamin C and E on cypermethrin -induced changes in serum potassium ion and chloride ion concentrations in female wistar rats

Cypermethrin resulted in significant increase in serum potassium and chloride ion concentrations in the exposed rats when compared to control, administration of vitamins C and E individually significantly adjusted the concentration of these ions (fig 2).

The effect of administration of vitamin C and E on cypermethrin -induced changes in serum bicarbonate and sodium ion concentrations in female wistar rats

Cypermethrin resulted in significant increase in serum bicarbonate and sodium ion concentrations in the exposed rats when compared to control. Exposure of these rats individually to vitamins C and E significantly modulated the concentration of these ions (fig 3).

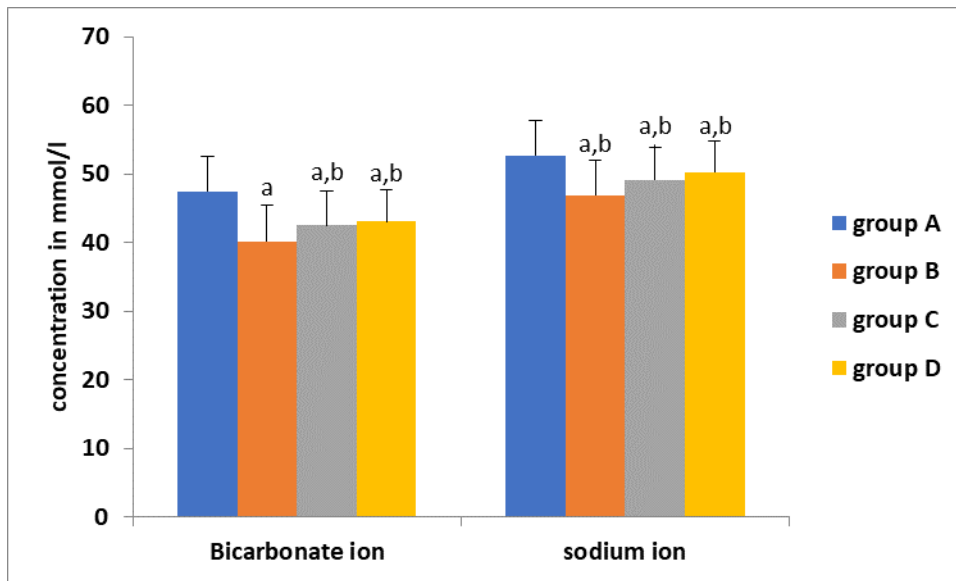


Fig 3. The effect of administration of vitamin C and E on cypermethrin -induced changes in serum bicarbonate and sodium ion concentrations in female wistar rats
 Values are means \pm SD. ^a values are significantly different from normal control (group A) at $P < 0.05$. while ^b values are Significantly different from cypermethrin group (group B) at $P < 0.05$

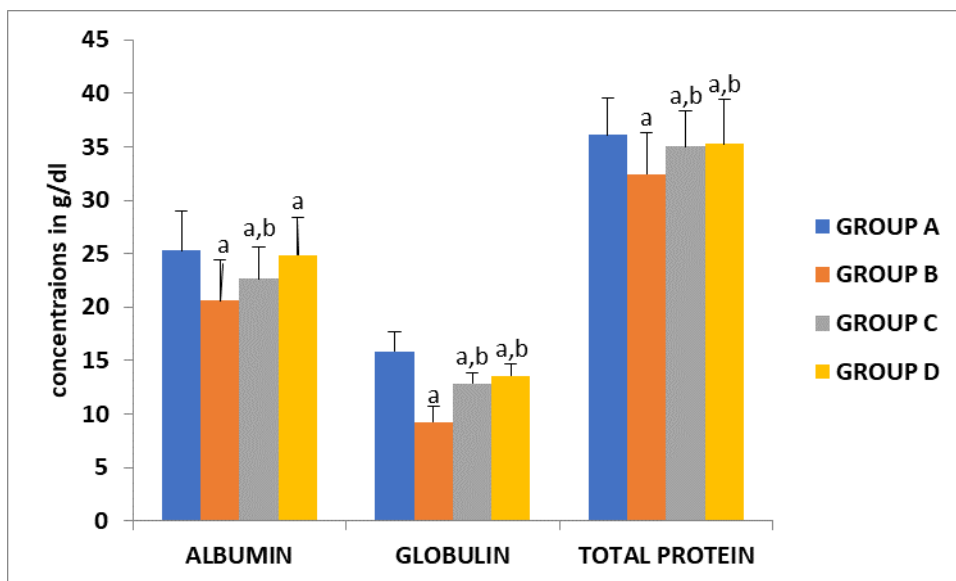


Fig 4. The effect of administration of vitamin C and E on cypermethrin -induced changes in renal albumin, globulin and total protein in female wistar rats
 Values are means \pm SD. ^a values are significantly different from normal control (group A) at $P < 0.05$. while ^b values are Significantly different from cypermethrin group (group B) at $P < 0.05$

The effect of administration of vitamin C and E on cypermethrin -induced changes in renal albumin, globulin and total protein in female wistar rats

Renal total protein, albumin and globulin concentrations were significantly reduced upon exposure to cypermethrin, administration of

vitamins C and E to these rats induced significant increase in the concentrations of these proteins (fig 4)

Effects of Vitamins C and E on histology of the kidney of rats exposed to cypermethrin

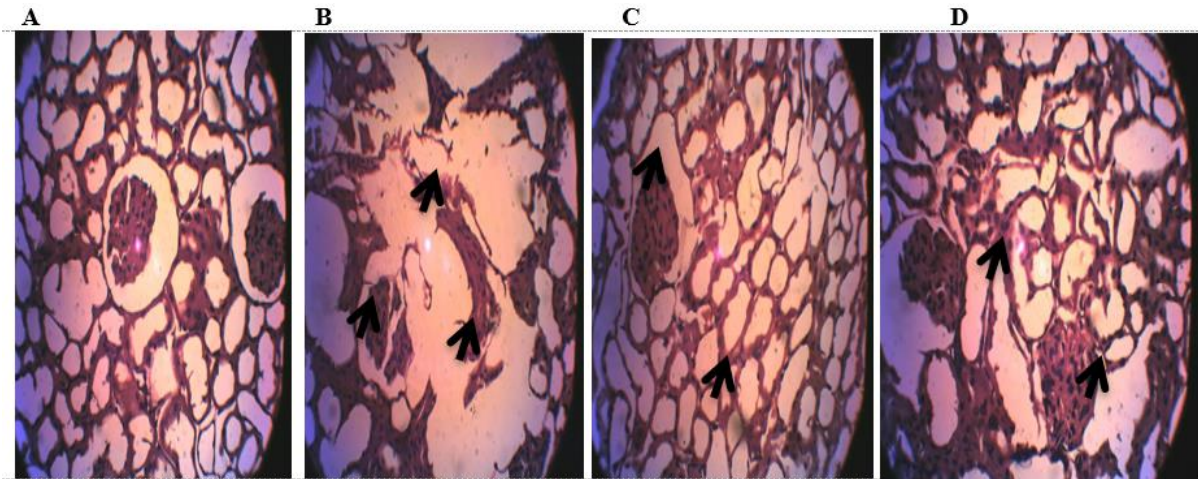


Fig. 5. Photomicrographs of section of renal tissues of rats administered Cypermethrin, vitamin C and E. A: control rats showing normal architecture of the renal tissue; **B:** rats administered cypermethrin only showing distorted histo architecture characterized by lesion of glomerulus, damaged Bowman's capsule, degenerated and vacuolated renal tubules (black arrows); **C:** rats administered cypermethrin + Vitamin C showing intact glomerulus, dilated capsular space and vacuolization of renal tubules with flattened cuboidal epithelium (black arrows); **D:** rats administered cypermethrin + Vitamin E showing dilated capsular space, intact glomerulus, degenerated and vacuolated renal tubules (black arrows).

Discussion

The observed significant increase ($P < 0.05$) in the level of urea, uric acid and creatinine in the serum of cypermethrin treated rats as compared to the normal control is an indication of renal dysfunction. Urea is indicator of renal function which is routinely measured to assess the kidney health status while Creatinine is an important marker of the filtration function of the kidneys because it is chiefly excreted from the blood via glomerular filtration. The increased level of these serum renal metabolites may have resulted from decreased in the rate at where they are excreted which may be as a result of impairment in kidney ability to carry out the biochemical process (Oyewole and Oladele, 2016; Oladele and Oyewole, 2017). However, administration of vitamin C and E significantly ameliorate the alterations in kidney functions confirming the role of the vitamin in protecting the integrity of membranes.

Administration of cypermethrin in this study significantly ($P < 0.05$) increased serum Potassium ion (K^+) and chloride ion levels; and significantly ($P < 0.05$) decreased serum Sodium ion (Na^+) and bicarbonate ions (HCO_3^-) causing electrolyte disturbance in the experiment animals (Oladele et al., 2017). Hyperchloremia is an electrolyte disturbance in which there is an abnormally elevated level of the chloride ion in the blood. Elevations in chloride may be associated with administration of significant amounts of IV normal saline, diarrhea, certain kidney diseases as type 2 renal tubular acidosis, type 1 renal tubular acidosis, and over activity of the parathyroid glands. Hyperchloremia is often comorbid with diabetes or

hyponatremia. Hyponatremia occurs because of an imbalance of water and sodium. Low sodium levels are uncommon and are most often caused by heart failure, malnutrition, and diarrhea.

Treatment with vitamin C and E was found to significantly reverse the alterations in the serum electrolyte homeostasis. This may be attributed to the ability of the vitamins to participate in the cell signalling pathways thus mitigating the biological processes that may result into electrolyte imbalance in the serum.

The results from this study reveal that there is a significant decrease in the renal total protein, albumin and globulin concentrations following administration of Cypermethrin. Total protein, albumin and globulin concentrations are an essential tool in assessing the renal toxicity profile of xenobiotics. Low levels of these proteins including albumin and globulin have been associated with kidney damage, malnutrition and dehydration (Oyewole et al., 2012). Globulin and albumin are globular proteins which are synthesized in the liver and transported by blood circulation thus found in the serum. Treatment with vitamin C and E significantly attenuate these toxic effects.

The histological result shows that the administration of cypermethrin shows distorted histoarchitecture characterised by lesion of glomerulus, damaged Bowman's capsule, degenerated and vacuolated renal tubules. The ameliorating effect of vitamin C and E was noticed as they both reduced the effect of cypermethrin on the kidney by making the glomerulus intact. This result corroborates the biochemical studies.

CONCLUSION

The present study demonstrated that exposure of rats to cypermethrin induced renal dysfunction and causes alteration in the serum electrolytes homeostasis with increase in serum chloride ion, potassium ion and decrease in serum level of sodium ion and bicarbonates. However, treatment with vitamin C and E offers protective actions against the toxic effects of cypermethrin.

Competing interests

The authors declare that they have no competing interests.

References

- Bell, E. F. (1987). History of vitamin E in infant nutrition. *American Journal of Clinical Nutrition*, 46 (1 Suppl): 183–186.
- Cheesbrough, M. (2005). Measurement of serum or plasma creatinine and urea. In, District laboratory practice in tropical countries. 2nd Edition. *Cambridge University Press*, 333-340.
- Das, B. K., & Mukherjee, S. C. (2003). Toxicity of cypermethrin in Labeorohita fingerlings: Biochemical enzymatic and haematological consequences. *comp. Biochem. Physiol.*, 134(C):109-121.
- Elliott, M., Farnham, A. W., Janes, N. F., Needham, P. H., & Pulman, D. A. (1974). Synthetic insecticide with a new order of activity. *Nature*, 248, 710–717.
- Gowland, B. T. G., Moffat, C. F., Stagg, R. M., Hovlihon, D. F., & Davies, I. M. (2002). Marine Cypermethrin induces glutathione s-transferase activities in the shore crab *Carcinusmaenas*, *Environ Res.*, 2002, 54,169-177
- Lowry, O. H., Rosebrough, N. J., Farr, A. L., & Randall, R. J. (1951). Protein measurement with the Folin phenol reagent. *J. Biol. Chem.*, 193, 265-275.
- Mokady, I. C., Abramovici, A., & Cogan, U. (1989). The safety evaluation of *dunaviela bardawilla* as a potential food supplements. *Food and Chemical Toxicology*, 27, 221-226.
- National Research Council (NRC) (2011). Guide for the care and use of laboratory animals 8th Edition. *The National Academies Press*.
- Oladele, J. O., Oyewole, O. I., Bello, O. K., Oladele, O. T. (2017). Modulatory properties of *Telfairia occidentalis* leaf extract on pancytopenia, electrolyte imbalance and renal oxidative damage in rats. *J. Biosci. Biotechnol. Discov.* 2: 74-78.
- Oyewole, O. I., & Oladele, J. O. (2016). Assessment of Cardiac and Renal Functions in Wistar Albino Rats Administered Carmoisine and Tartrazine. *Advances in Biochemistry*. 4, 3, 21-25.
- Oyewole, O. I., and Oladele, J. O. (2017). Changes in activities of tissues enzymes in rats administered *Ficus exasperata* leaf extract. *Int. J. Biol. Chem. Sci.* 11(1): 378-386.
- Oyewole, O. I., Dere, F. A., & Okoro, O. E. (2012). Sodium benzoate mediated hepatorenal toxicity in Wistar Rat: Modulatory effects of *Azadirachta indica* (Neem) leaf. *European Journal of Medicinal Plants*. 2(1): 11-18.
- Tietz, N. W., Pruden, E. L., & Siggaard-Anderson, O. (1994). In: *Tietz textbook of Clinical Chemistry* (Burtis CA, Ashwell ER eds.) WB Saunder Co. London. pp. 1354-1374.
- Traber, M. G., Stevens, J. F. (2011). Free Radical Biology and Medicine – Vitamins C and E: Beneficial effects from a mechanistic perspective. *Free Radical Biology and Medicine*. 51 (5): 1000–1013.
- Usmani, K. A., & Knowles, C. O. (2001). Toxicity of pyrethroids and effect of synergists to: larval and adult *Helicoverpa Zea*, *Spodopterafrugiperda*, and *Agrotis ipsilon* (Lepidoptera Noctuidae). *J. Econ. Entomol.*, 94: 868–73.