British Journal of Pharmaceutical Research
6(5): 293-305, 2015, Article no.BJPR.2015.071
ISSN: 2231-2919

SCIENCEDOMAIN *international*
www.sciencedomain.org

Alterations in Biochemical Indices and Antioxidant Status in Rats Following Treatment with Gatifloxacin

Ebenezer Tunde Olayinka[1*], Ayokanmi Ore[1] and Oluwatobi Adewumi Adeyemo[1]

[1]Department of Chemical Sciences, Biochemistry Unit, P.M.B.1066, Ajayi Crowther University, Oyo, Nigeria.

Authors' contributions

This research work was carried out in collaboration between all the authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/BJPR/2015/16210
Editor(s):
(1) Jinyong Peng, College of Pharmacy, Dalian Medical University, Dalian, China.
Reviewers:
(1) Li Xing, Davison of Biomedical Statistics and Informatics and Department of Health Sciences Research, Mayo Clinic, USA.
(2) Anonymous, Slovenia.
Complete Peer review History: http://www.sciencedomain.org/review-history.php?iid=986&id=14&aid=8658

Original Research Article

Received 15th January 2015
Accepted 11th March 2015
Published 1st April 2015

ABSTRACT

Aim: Gatifloxacin (GTX) - an 8-methoxy fluoroquinolone antibacterial agent has been considered very effective in the treatment of respiratory and urinary tract infections. This study investigates the toxic potentials of GTX in Wistar rats.
Methodology: Twenty male rats (180-220 g) were randomised into four groups: I-Control, II-4 mg/kg body weight (b.w.) GTX, III-8 mg/kg b.w GTX, and IV-16 mg/kg b.w GTX.
Results: After seven days of GTX administration, the levels of plasma creatinine, urea and bilirubin were increased significantly ($P<0.05$) in GTX-treated rats compared to control. ALP, ALT, AST and GGT activities were also elevated significantly in the plasma of the treated animals relative to control. Similarly, hepatic malondialdehyde level increased significantly in the GTX-treated groups relative to control. Hepatic levels of ascorbic acid, reduced glutathione as well as activities of hepatic GST, catalase, and SOD were reduced significantly in a dose dependent manner in the GTX-treated animals compared to control. Besides, histopathological studies revealed very mild, moderate and severe hepatic portal congestion and cellular infiltration by mononuclear cells by the three doses of GTX.
Conclusion: Overall, three different doses of Gatifloxacin (half-therapeutic, therapeutic and double-therapeutic) induced renal and hepatic damages, as well as oxidative stress in rats.

**Corresponding author: Email: ebenezertundeolayinka@gmail.com, et.olayinka@acu.edu.ng;*

Keywords: Gatifloxacin; liver damages; oxidative stress; half-therapeutic; therapeutic; double-therapeutic doses.

1. INTRODUCTION

The fluoroquinolones are a family of broad spectrum, systemic antibacterial agents that have been used widely for the treatment of respiratory and urinary tract infections. Gatifloxacin (1-cyclopropyl-6-fluoro-8-methoxy-7 (-3-methyl piperazin-1-yl)-4-oxo-quinoline-3-carboxylic acid) (Fig. 1), is an antibiotic of the fourth-generation fluoroquinolone family. Like other members of that family, gatifloxacin (GTX) inhibits the bacteria enzyme DNA gyrase and topoisomerase IV [1]. GTX is active against gram positive and gram negative organisms including anaerobes such as mycoplasma, chlamydia, legionella and mycobacteria [2,3]. GTX also penetrates well into leukocytes which can deliver active drug to sites of infection and plays important role in the treatment of intracellular pathogens [3].

The fluoroquinolone antibacterial agents such as morfloxacin, ofloxacin, ciprofloxacin and lomefloxacin possess photosensitizing properties leading to phototoxic responses [4]. Several *in vivo* and *in vitro* studies have demonstrated that there is a relationship between phototoxicity and generation of reactive oxygen species (ROS) [5,6,7]. Studies have also shown that ciprofloxacin and levofloxacin induced oxidative stress and lipid peroxidation in humans and rats by generating ROS [8,9]. Oxidative stress arises when there is an imbalance between the generations of ROS and the activity of the antioxidant defenses. Normally, a state of equilibrium exists between tissue oxidants and antioxidant activities [10,11]. This balance can however, be upset as a result of excessive free radical generation, depletion of endogenous antioxidants or failure to repair oxidative injury by ROS [12]. There are antioxidant defense systems protecting against the damages caused by ROS in the body, thus maintaining redox homeostasis. These include enzymatic (catalase, superoxide dismutase (SOD), glutathione peroxidase etc) and non- enzymatic (vitamin A, C, E and glutathione) antioxidants. Oxidative stress emanating from generation of ROS has been implicated in lipid peroxidation, protein modification and DNA damage resulting in pathogenesis of certain diseases such as aging, cancer, cardiovascular disorder, neurodegenerative diseases etc [13,14,15].

Gatifloxacin has been found to be associated with increased risk of dysglycemia (hypoglycemia and hyperglycemia) in humans [16,17]. In addition, it has been reported that other fluoroquinolone antibiotics generate ROS and cause oxidative stress [8,9]. The present study was therefore designed to evaluate the influence of three different doses of Gatifloxacin on some biochemical parameters and antioxidant status in rats.

2. MATERIALS AND METHODS

2.1 Chemicals and Reagents

Gatifloxacin (OGAT®) is a product of Elegant drugs PVT Ltd, Chalmatti, Karnataka, India. 1-Chloro-2,4-dinitrobenzene (CDNB), 5,5'-dithio-bis-2-nitrobenzoic acid (DTNB), Glutathione (GSH), epinephrine, hydrogen peroxide (H_2O_2) and Thiobarbituric acid (TBA) were all purchased from Sigma Chemical Company (London, UK). Kits for Alanine aminotransferase (ALT), Aspartate aminotransferase (AST), Alkaline phosphatase (ALP), Urea, Creatinine, Bilirubin, and L-γ-Glutamyltransferase (GGT) were obtained from Randox Laboratories Ltd (Antrim, UK). All other chemicals and reagents used were of analytical grade and of highest purity.

2.2 Animals

In bred 5-6 weeks-old male rats (Wistar strain) weighing 180-220g were used in this study. The rats were bred and housed in the animal house of the Department of Chemical Sciences, Ajayi Crowther University, Oyo, Nigeria. They were kept in wire meshed cages at room temp (28-30°C) and under controlled light cycle (12-hr light: dark).They were fed with commercial rat chow (Ladokun feeds, Ibadan, Nigeria) and supplied water *ad libitum*. This study was approved by Animal Care and Use Committee, a sub-committee of the Ethical review committee, Faculty of Natural Sciences, Ajayi Crowther University. Handling of experimental animals conformed to the international guide on the care and use of laboratory animals *(National Research Council, 2011)*.

2.3 Treatments

Twenty (20) healthy male albino rats (Wistar strain) were randomly divided into four groups of

five (5) rats each. Group I (Control) received physiological saline, while groups II (GTX 1), III (GTX2) and IV (GTX3) were administered 4 mg/kg b.w. of Gatifloxacin (half-therapeutic dose), 8 mg/kg b.w. of Gatifloxacin (therapeutic dose) and 16 mg/kg b.w. of Gatifloxacin (double-therapeutic dose) respectively as presented in Table 1. 1 ml each of the prepared GTX solution was administered orally, once daily for seven days using an oral cannula.

Table 1. Experimental design

Experimental groups	Treatments
Group I *(n=5)*	Control (Normal saline)
Group II *(n=5)*	GTX 1 (4 mg/kg.b.w Gatifloxacin)
Group III *(n=5)*	GTX 2 (8 mg/kg.b.w Gatifloxacin)
Group IV *(n=5)*	GTX 3 (16 mg/kg. b.w Gatifloxacin)

2.4 Place and Duration of Study

This research was carried out in the Biochemistry unit, Department of Chemical Sciences, Faculty of Natural Sciences, Ajayi Crowther University, Oyo, Nigeria between April and June, 2014.

2.5 Collection of Blood Samples for Plasma Preparation

Blood was collected from the retro-orbital plexus of the animals into heparinized tubes, and animals were euthanized 24 hours after the last treatment. Plasma was prepared by centrifuging blood samples for ten minutes at 3000 rpm in a CENCOM® bench centrifuge. The clear supernatant was used for the estimation of urea, creatinine, bilirubin, and enzymes.

2.6 Preparation of Cytosolic Fractions

The liver, excised from rat, blotted of blood stains, rinsed in 1.15% KCl was homogenized in 4 volumes of ice-cold 0.01 M potassium phosphate buffer, (pH 7.4). The homogenates were centrifuged at 12,500 g in an Eppendorf (UK) refrigerated centrifuge for 15 min at 4°C and the supernatants, termed the post-mitochondrial fractions (PMF) were aliquoted and used for enzyme assays.

2.7 Renal and Liver Functions Test

Plasma creatinine, urea and bilirubin determination was done using Randox diagnostic kits. Methods for creatinine assays are based on colorimetric alkaline picrate methods of Jaffe [18] with Creatinine-picrate complex measured at 492 nm. The urea determination method was based on the fearon reaction [19], with the Diazine chromogen formed absorbing strongly at 540 nm. The dimethy sulphoxide method by Tietz et al. [19] was used for bilirubin determination. The dimethyl sulphoxide form a coloured compound with maximum absorption at 550 nm.

2.8 Determination of Plasma AST, ALT, ALP and GGT Activities

Plasma alanine amino transferase (ALT), aspartate amino transferase (AST), alkaline phosphatase (ALP) and gamma glutamyl transferase (GGT) activities were determined using Randox diagnostic kits. Determination of AST and ALT activities were based on the principle described by Reltman and Frankel [20]. AST was measured by monitoring the concentration of oxaloacetate hydrazone formed with 2,4-dinitrophenylhydrazine at 546nm and ALT was measured by monitoring the concentration of pyruvate hydrazone formed with 2,4-dinitrophenylhydrazine at 546 nm. ALP was determined in accordance with the principles of Tietz [21]. The p-nitrophenol formed by the hydrolysis of p-Nitrophenyl phosphate confers yellowish colour on the reaction mixture and its intensity can be monitored at 405 nm to give a measure of enzyme activity. GGT activity was measured based on a modification of the method described by Theodorsen et al. [22] using Abbott diagnostic kit.

2.9 Assay of Non-enzymatic Antioxidants and Lipid Peroxidation

Hepatic vitamin C was determined chemically according to the method of Erel et al. [23] using dinitro phenyl hydrazine (DNPH), while hepatic glutathione was determined according to the method of Jollow et al. [24]. The chromophoric product resulting from the reaction of Ellman's reagent with the reduced glutathione, 2-nitro-5-thiobenzoic acid possesses a molar absorption at 412 nm which was read in a spectrophotometer. Reduced GSH is proportional to the absorbance at 412 nm. The extent of lipid peroxidation (LPO) was estimated by the method of Vashney and Kale [25], the method involved the reaction between malondialdehyde (MDA; product of LPO) and thiobarbituric acid to yield a stable pink chromophore with maximum absorption at 532 nm.

2.10 Determination of Antioxidant Enzymes

The procedure of Misra and Fridovich [26] as described by Magwere et al. [27] was used for the determination of hepatic superoxide dismutase (SOD) activity by measuring the inhibition of auto-oxidation of epinephrine at pH 10.2 and 30ºC. Hepatic catalase activity was determined according to the method of Sinha [28] by measuring the reduction of dichromate in acetic acid to chromic acetate at 570 nm. Hepatic Glutathione S-transferase (GST) activity was determined by the method described by Habig et al. [29] using 1-chloro-2,4-dinitrobenzene (CDNB) as substrate.

2.11 Protein Determination

Protein determination of plasma and all fractions was estimated by the method of Lowry et al. [30] using bovine serum albumin as standard.

2.12 Histopatological Studies

The method of Baker and Silverton [31] was employed for the processing of liver for histopathological studies.

2.13 Statistical Analysis

The results were expressed as mean of 5 replicates ± SD. Data obtained were subjected to one-way analysis of variance (ANOVA) followed by Duncan's multiple range test to identify significantly different groups using SigmaPlot® Statistical Software. A value of $P < 0.05$ was accepted as statistically significant.

3. RESULTS

3.1 Effect of GTX on Plasma Creatinine, Urea and Bilirubin Level in Rats

The effect of GTX on plasma creatinine, urea and bilirubin level are shown in Table 2. Treatment with GTX 1, 2 and 3 significantly *(P < 0.05)* increased plasma creatinine, urea and bilirubin level by 21.2%, 63.2%, 106.1%; 24%, 55.5%, 96%; 46.9%, 95.2% and 163.9% respectively when compared with the control value.

3.2 Effect of GTX on the Activities of Plasma Enzymes in Rats

Table 3 presents the effect of GTX on plasma ALP, ALT, AST and GGT activities in rats. The activities of ALP, ALT, AST and GGT were significantly increased in the plasma of treated rats by 43.5%, 117.1%, 157.2%; 31.5%, 52.8%, 95.4%; 19.7%, 44.6%, 56.1%; and 25.7%, 58.6%, 81.4% respectively relative to the control.

3.3 Effect of GTX on the Activities of Antioxidant Enzymes in Rat Liver

Table 4 shows the effect of GTX treatment on hepatic superoxide dismutase (SOD) and catalase (CAT) activities in rats. GTX 1, 2 and 3 treatment significantly decrease the activities of SOD and CAT in the liver of rats by 10.6%, 14.5%, 21.6%; 30.4%, 40.7% and 56.5% respectively when compared with control *(P < 0.05)*. Also, treatment with GTX 1, 2 and 3 reduced significantly the activity of hepatic glutathione –S- transferase (GST) by 14%, 30.6% and 43.4% respectively relative to the control (Fig. 3).

3.4 Effect of GTX on the Levels of Hepatic Non-enzymatic Antioxidant and Lipid Peroxidation in Rats

The hepatic Vitamin C concentration is shown in Fig. 4 following treatment with GTX (GTX 1, 2, 3). The Vitamin C level was significantly decreased by 9.7%, 20.9% and 35.6% respectively in the treated groups when compared with the control. Also, hepatic GSH level was significantly decreased by 31.9%, 48.2% and 69.2% respectively in the treated

Table 2. Effect of gatifloxacin on plasma creatinine, urea and bilirubin levels in rats

Treatment	Creatinine (mg/dl)	Urea (mg/dl)	Bilirubin (mg/dl)
Control	0.42±0.02	11.±0.4	0.29±0.01
GTX 1	0.51±0.01 (21.2%)*	13.7±0.1 (24%)*	0.43±0.01 (46.9%)*
GTX 2	0.72±0.02 (63.2%)*	17.1±0.12 (55.5%)*	0.57±0.01 (95.2%)*
GTX 3	0.87±0.04 (106.1%)*	21.6±1.1 (96%)*	0.77±0.01 (163.9%)*

*The values are the Means ± SD for five rats in each group, * significantly different from the control p<0.05, values in parenthesis represent percentage (%) increase*

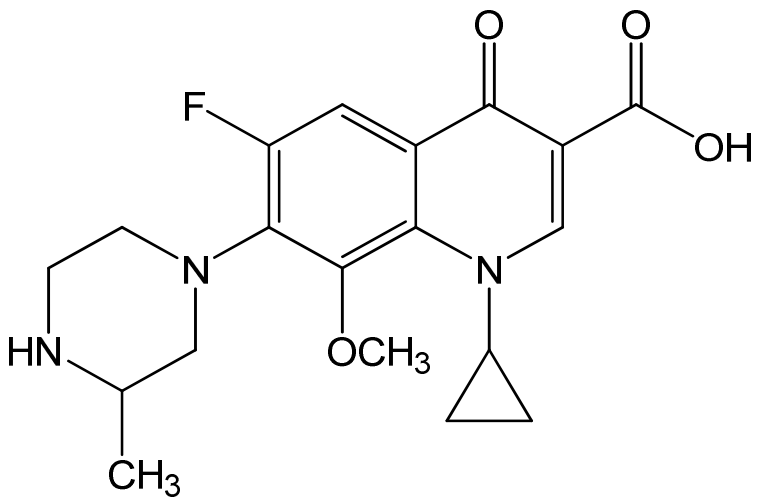


Fig. 1. Structure of gatifloxacin

Table 3. Effect of gatifloxacin on plasma alkaline phosphatase (ALP), aspartate amino transferase (AST), alanine amino transferase (ALT) and gamma glutamyl transferase (γ-GT) activities in rats

Treatment	ALP (U/L)	ALT (U/L)	AST (U/L)	GGT (U/L)
Control	285.0±4.3	21.6±1.1	61.0±1	14.0±1.6
GTX 1	409.0±8 (43.5%)*	28.4±1.5 (31.5%)*	73.0±1.6 (19.7%)*	17.6±1.1 (25.7%)*
GTX 2	619.0±4.2 (117.1%)*	33.0±1.6 (52.8%)*	88.2±1.8 (44.6%)*	22.2±0.8 (58.6%)*
GTX 3	734.0±2.6 (157.2%)*	42.2±1.9 (95.4%)*	95.2±1.9 (56.1%)*	25.4±1.1 (81.4%)*

*The values are the means ± SD for five rats in each group, * significantly different from the control p<0.05, values in parenthesis represent percentage (%) increase*

Table 4. Effect of gatifloxacin on hepatic catalase (CAT) and superoxide dismutase (SOD) activities in rats

Treatment	Catalase (μmole H_2O_2 consumed/min)	SOD (Unit)
Control	102.0±1.6	9.2±0.6
GTX 1	91.2±1.3 (10.6%)*	6.4±0.4 (30.4%)*
GTX 2	87.2±1.6 (14.5%)*	5.5±0.5 (40.7%)*
GTX 3	80.0±1.8 (21.6%)*	4.0±0.4 (56.5%)*

*The values are the means ± SD for five rats in each group, * significantly different from the control p<0.05, values in parenthesis represent percentage (%) decrease*

groups relative to the control (Fig. 2). However, hepatic lipid peroxidation (MDA) level was significantly increased by 17.5%, 57.3% and 92.3% in the treated groups respectively when compared with the control (Fig. 5).

3.5 Effect of GTX on the Histopathology of Liver of Rats

Fig. 6 is a photomicrographs showing the effect of GTX (GTX 1, 2 and 3) on the liver of rats. There was very mild periportal degeneration of vacuoles and infiltration by mononuclear cells in the liver of rats treated with GXT 1 (Fig. 6B). The picture also indicate moderate periportal degeneration of vacuole and mild periportal infiltration of mononuclear cells in the liver of GTX 2 treated rats (Fig. 6C). Moreover, severe portal congestion and mild periportal infiltration by mononuclear cells was observed in the liver of GTX 3 treated rats (Fig. 6D).

4. DISCUSSION

Fluoroquinolones have been shown to generate reactive oxygen species (ROS), as well inducing oxidative stress in human and experimental animals [8]. Studies have also shown that the

generation of ROS by fluoroquinolones resulted in cellular damage to the liver and kidney [32,33,34].

The potential toxicity of three doses of GTX in the liver and kidney together with effect on antioxidant status was investigated in rats. Our results indicated that administration of GTX induced marked renal and liver injury and depleted both the enzymatic and non-enzymatic antioxidant system. Regulation of antioxidant enzyme in tissues of higher animals is dependent on variety of factors and their concentrations in the cell is rapidly altered in response to transcriptional regulators as a result of sudden changes in oxidant levels [35]. Enzyme and metabolite biomarkers are suitable indicators for the diagnosis of tissue damages occationed by administration of drugs [36].

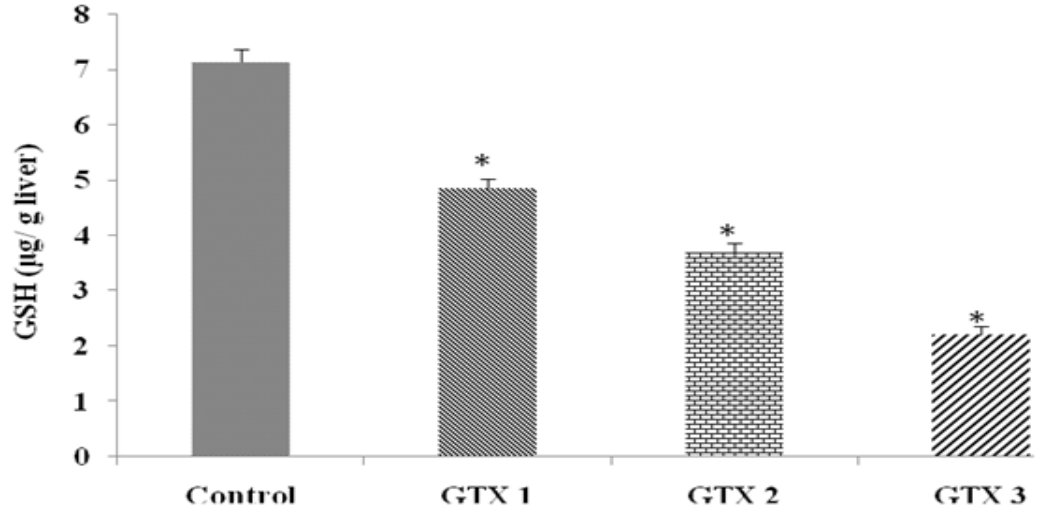


Fig. 2. Effect of gatifloxacin on hepatic glutathione (GSH) concentration in rats
*The values are the means ± SD) for five rats in each group, * significantly different from the control (P<0.05)*

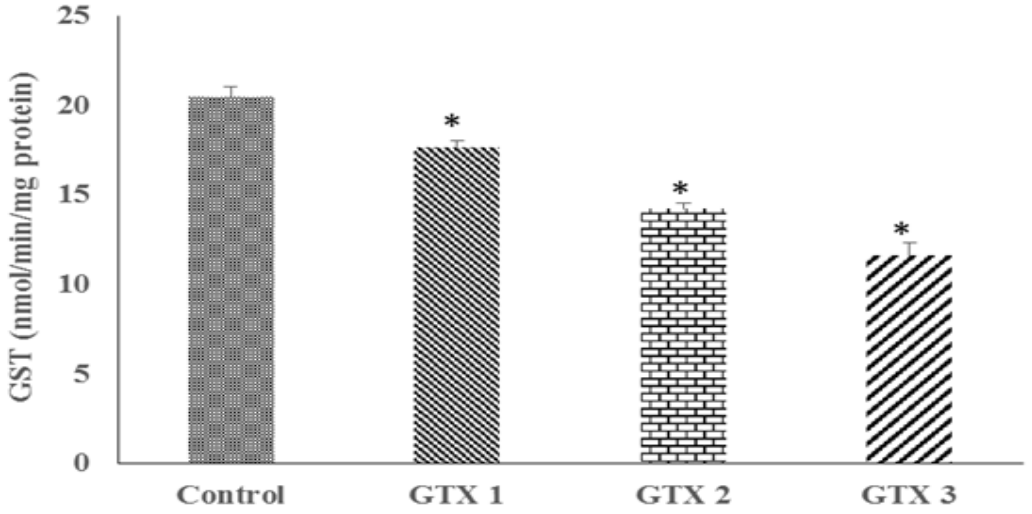


Fig. 3. Effect of gatifloxacin on hepatic glutathione S-transferase (GST) activity in rats
*The values are the means ± SD for five rats in each group, * significantly different from the control (P<0.05)*

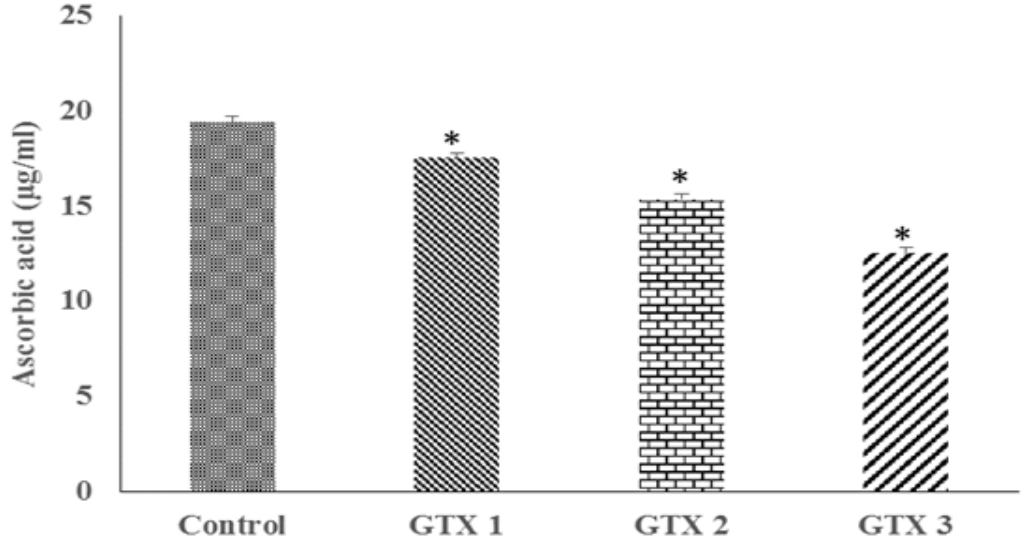


Fig. 4. Effect of gatifloxacin on hepatic ascorbic acid concentration in rats
*The values are the means ± SD for five rats in each group, * significantly different from the control (P<0.05)*

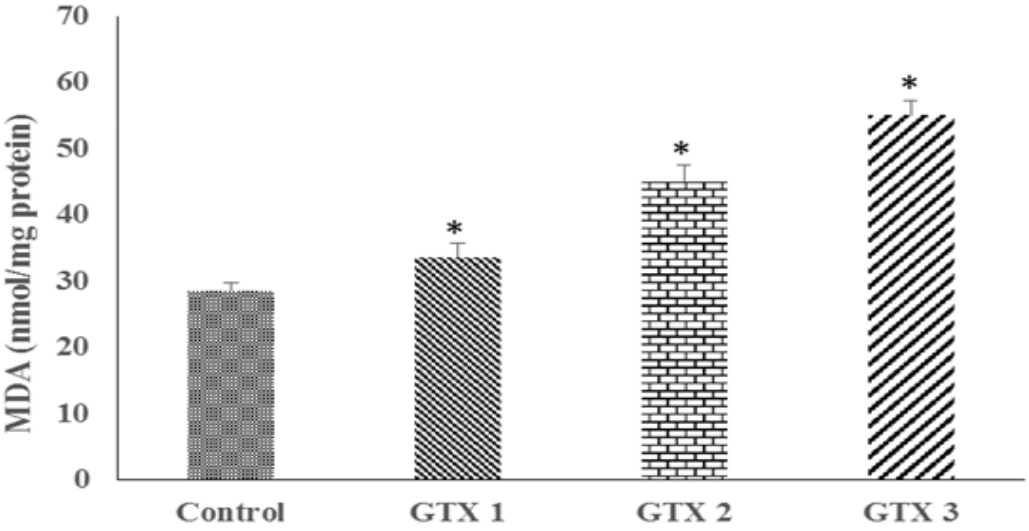
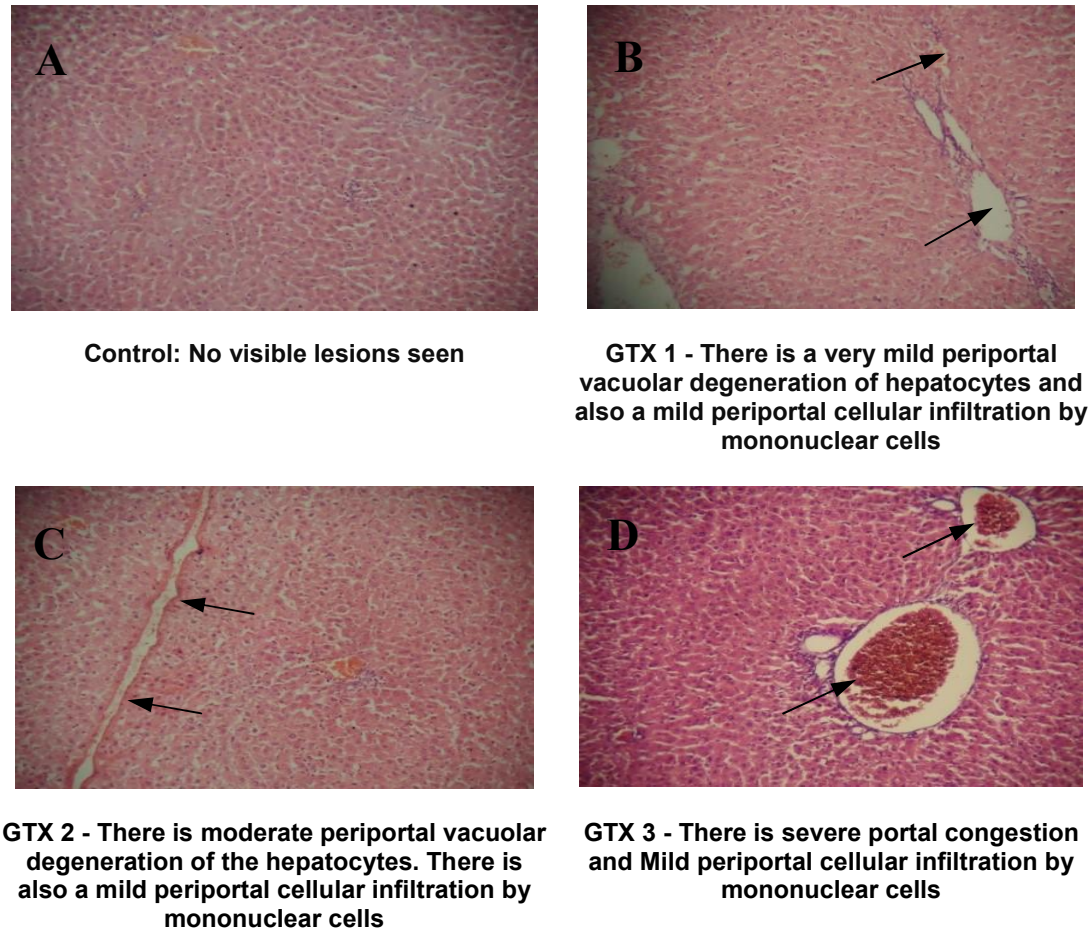


Fig. 5. Effect of gatifloxacin on hepatic lipid peroxidation (LPO) in rats
*The values are the means ± SD for five rats in each group, *significantly different from the control (P<0.05)*

The enzymic antioxidant defense system are the natural protector against lipid peroxidation. SOD and CAT are important scanvengers of superoxide ion and hydrogen peroxide. The two enzymes together with glutathione peroxidase prevent the generation of hydroxyl radical and protect cellular constituents from oxidative damage [37]. SOD is involved in the rapid dismutation of superoxide anion (O_2^-) to hydrogen peroxide (H_2O_2). The H_2O_2 generated from the dismutation is also a powerful membrane penetrating oxidant which has to be rapidly removed from the cell [38]. The elimination of H_2O_2 in the cell can either be handled by catalase (CAT) or glutathione peroxidase [39,40,41] in reactions that produces water and molecular oxygen.

The increased production of ROS occasioned by exposure of the animals to three doses of GTX may have resulted in the reduction of SOD and CAT activities in the liver. Similar result was



Control: No visible lesions seen

GTX 1 - There is a very mild periportal vacuolar degeneration of hepatocytes and also a mild periportal cellular infiltration by mononuclear cells

GTX 2 - There is moderate periportal vacuolar degeneration of the hepatocytes. There is also a mild periportal cellular infiltration by mononuclear cells

GTX 3 - There is severe portal congestion and Mild periportal cellular infiltration by mononuclear cells

Fig. 6. Photomicrograph of the liver showing the effect of the antibiotics gatifloxacin at different doses

reported by other researchers [8,42]. Administration of GTX may have induced adaptive responses including a derangement of some hepatocyte antioxidant mechanism (decreased SOD and CAT activities).

Glutathione-S-transferase (GST) is a group of multigene isozymes found in prokaryotes and eukaryotes best known for their ability to catalyse the conjugation of the reduced form of glutathione to xenobiotic substrate [43], including detoxification of drugs and harmful free radical by- products of metabolism resulting in elimination of toxic compounds [44-46]. The enzyme has also been found to have antioxidant properties [47-49]. The inhibition of GST activity by the three doses of GTX may have some implications; including the capacity of the liver to detoxify xenobiotics possibly being impaired by the drug [36].

Reduced glutathione (GSH) is the most important non enzymatic antioxidant defence [50]. The antioxidant effect of GSH can be expressed in different ways [51]; detoxification of hydrogen peroxide and lipid peroxides catalysed by glutathione peroxidase (GSH-Px); it donates its electron to H_2O_2 to reduce it into H_2O and O_2 [52]; serving as a cofactor for several detoxifying enzymes, such as GSH-Px and transferase and has a role in converting vitamin C and E back to their active forms [50]. In higher animals, ascorbic acid (vitamin C) in addition to vitamin A and E are known to represent the first line of antioxidant defence [53,54] and this vitamin is likely to be most susceptible to free radical oxidation. Ascorbate is a good free radical scavenger due to its chemical properties [55]. Several studies have reported that there are significant interrelationship between the intracellular concentration of vitamin C and GSH

and their ability to mutually spare each other [41,56]. The significant reduction in the hepatic vitamin C and GSH concentration by GTX is an indication of decrease in the overall redox status of the liver, emanating from the formation of reactive oxygen species or toxic metabolites by the three doses of GTX.

Oxidative damage to unsaturated lipids is a well-established general mechanism for oxidant mediated cellular injury [57]. In addition to extensive experimental studies, increased lipid peroxidation has been reported in a wide variety of clinical and toxicological conditions [58]. Free radicals may damage the liver through mechanisms of covalent binding and lipid peroxidation with subsequent tissue injury [59]. Malondialdehyde (MDA), which is one of the products of lipid peroxidation, has been the most extensively studied marker. Increased MDA is an indication of ROS generation in the tissue and increase in the level of lipid peroxidation has been described as a biomarkers of tissue damage [60]. The apparent increase in MDA formation indicates that GTX is capable of inducing oxidative stress which may overload the cells endogenous detoxification mechanism. This observation agrees with previous studies on certain fluoroquinolones antibiotics [9].

The liver is a primary organ involved in the metabolism and excretion of toxic substances including drugs from the body. Liver damages are mainly caused by an imbalance in the accumulation of toxicant and the drug metabolizing ability of the liver [61,62]. Considering the complex and diverse nature of the functions of the liver, there are array of tests being employed in measuring the extent of liver damage [63,64]. The magnitude of derangement of liver by hepatotoxicant is generally measured by the level of ALT, AST, ALP and bilirubin [65].

ALT, AST and ALP are marker enzymes for hepatocellular damages [66]. There is an association between distortion of the membrane of liver cells and leakage of its cytosolic content. This is manifested by the significant increase in the level of the marker enzymes in the blood. The serum ALT has been regarded as a reliable and sensitive marker of liver disease. ALT in most organs including liver, heart, skeletal muscle, kidney, brain and pancreas. Owing to its wide tissue distribution, elevated AST have low specificity for any single disease. ALP are found primarily in the liver, bone, intestine, placenta, and the kidney. Elevated serum level of these marker enzymes gave an indication of tissue injury resulting in their leakage into the blood [67]. It has been reported that increase in plasma bilirubin maybe as a result of damages to liver cell, impairment in the uptake of unconjugated bilirubin in the liver, intra and extra-hepatic biliary tract obstruction, and neonatal jaundice [68,69,70]. The elevated aminotransferases, phosphatases and bilirubin observed in the three different doses of GTX-treated animals is highly indicative of hepatic toxicity [71,72].

The kidney is an organ mainly involved in drug elimination. It appears that the kidney is less able to protect itself from the effects of the radical species when compared to the liver that is known to be well equipped with several detoxifying enzymes. This could have serious implication on the renal functions of the animals on long-term prophylaxis [36]. This was evident in the significantly elevated levels of urea and creatinine in the plasma of rats following administration of GTX, suggesting an impairment of kidney function. Urea, a product of protein metabolism and creatinine, a product of creatine and phosphocreatine are excreted by the kidney [73]. Their plasma concentration are suitable indicator of kidney and cardiovascular functions [74,75]. Increase in the plasma levels of creatinine and urea observed in this study suggests an abnormal renal function [76,77].

The histological alteration observed in the liver of GTX-treated rats characterized by mild periportal degeneration of liver cell vacuole, moderate periportal degeneration of liver cell vacuole and severe portal congestion and mild periportal cellular infiltration by mononuclear cells may be a consequence of the formation of a highly reactive radicals owing to oxidative stress occasioned by the antibiotics, which disrupted normal cellular functioning of the liver.

5. CONCLUSION

Our results indicate that gatifloxacin altered the antioxidant status and induced kidney and liver damage. We have demonstrated a dose dependent alteration in most parameters measured. Data from the present study coupled with increased frequency of hypoglycaemia and hyperglycaemia might justify the call for withdrawal of gatifloxacin from market in some countries.

CONSENT

It is not applicable.

ACKNOWLEDGEMENTS

The research is a self-funded study and therefore the acknowledgement section can be left out.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Emmanuelle C, Stephanie M, Xiao-Su P, Romain RDB, Ce´line C, Alexandra A, Christine Lascols, Jean- Yves Driot and L. Mark Fisher. Target specificity of the new fluoroquinolone besifloxacin in *Streptococcus pneumoniae*, *Staphylococcus aureus* and *Escherichia coli*. J Antimicrob. Chemother. 2009;63: 443–450.
2. Zhao X, Wang JY, Xu C, et al. Killing of *Staphylococcus aureus* by C-8 methoxy fluoroquinolones. Antimicrob Agents Chemother. 1998;42:956-8.
3. Kumar NS, Dhivya D, Vijayakumar B. A focus on quinolones and its medicinal importance. Inter. J Novel Trends Pharm. Sci. 2011;1(1):23-29.
4. Martinez LJ, Sik RH, Chignell CF, Fluoroquinolone antimicrobials: Singlet oxygen, superoxide and phototoxicity in mice. Photochem. Photobiol. 1998; 65:495-9.
5. Fujita H, Matsuo I. Participation of re;active oxygen spiecies in phototoxicity induced by quinolone antibacterial agents. Photodermatol. Photoimmunol. Photomed. 1994;10(5):202-5.
6. Umezawa N, Arakane K, Ryu A, Mashiko S, Hirobe M, Nagano T. Participation of reactive oxygen species in phototoxicity induced by quinolone antibacterial agents. Arch. Biochem. Biophys. 1997;34(2):275-81.
7. Kawada A, Hatanaka K, Gomi H, Matsuo I. *Invivo and In vitro* phototoxic activities of new quinolone antibacterial agents;lipid peroxidative potential. Photodermatol photoimmunol photomed. 1999;15(6):226-30.
8. Talla V, Veerareddy PR. Oxidative stress induced by fluoroquinolones on treatment for complicated urinary tract infections in Indian patients. J. Young Pharm. 2011; 3(4):304–309.
9. Afolabi OK, Oyewo EB. Effects of Ciprofloxacin and Levofloxacin Administration on Some Oxidative Stress Markers in the Rat. Intl. Journal of Biol. Vet. Agric. Food Eng. 2014;8(1):80-84.
10. Sharma P, Jha AB, Dubey RS, Pessarakli M. Reactive Oxygen Species, Oxidative Damage, and Antioxidative Defense Mechanism in Plants under Stressful Conditions J. Botany. 2012;1-26.
11. Aly DG, Shahin RS, Oxidative stress in lichen planus. Acta Dermatovenerologica Alp. Pannonica et Adriatica. 2010;19(1):3-11.
12. Grigorov B. Reactive oxygen species and their relation to carcinogenesis. Trakia J of Sci. 2012;10(3):83-92.
13. Aruoma OI. Free radicals, oxidative stress, and antioxidants in human health and disease. J. Agric. Food Chem. 1999;47: 397-492.
14. Abuja PM, Albertini R. Methods for monitoring oxidative stress, lipid peroxidation and oxidation resistance of lipoproteins. Clinica. Chimica. Acta. 2001; 306:1–17.
15. Gábor C, Francis JM Jr. Oxidative stress in cardiovascular disease. Int. J. Mol. Sci. 2014;15:6002-6008.
16. Park-Wyllie LY, Juurlink DN, Kopp A Shah BR, Stukel TA, Stumpo C, Dresser L, Low DE, Mamdani MM. Outpatient gatifloxacin therapy and dysglycemia in older adults. N Engl J Med. 2006;354(13):1352-61.
17. Pichichero ME, Arguedas A, Dagan R, Sher L. Safety and efficacy of gatifloxacin therapy for children with recurrent acute otitis media (AOM) and/or AOM treatment failure. CID. 2005;41:470–8.
18. Jaffe, B. What made the radical break. N. Engl. J. Med. 1972;286:156-157.
19. Tietz NW, Pruden EL, Siaggard-Andersen O. In: Tietz textbook Clinical Chemistry (Burtis C.A and Ashwell E.R.eds) W.B Saunders Company London 1994;1354-1374.
20. Reitman S, Frankel S. Determination of plasma Amino Transferases Activities. Am. J Clin. Path. 1957;28:56.

21. Tietz, NW. Clinical guide to laboratory tests. 3rd edn., W. B. Saunders Company, Philadelphia; 1995.
22. Theodorsen L, Strömme JH, Hørder M, et al. Recommended method for the determination of γ-glutamyltransferase in blood. Scand J Clin. Lab. Invest. 1976; 36:711-723.
23. Erel, D, Kocyigit A, Avci S, Aktepe N, Bulut V. Oxidative stress and antioxidative status of plasma and erythrocytes in patients with malaria. Clin. Biochem. 1997;30:631-639.
24. Jollow, DJ, Mitchell JR, Zampahone N, Gillet JR. Bromobenzene induced liver necrosis; Protective role of glutathione and evidence for 3,4 Pharmacol. 1974;11:151-169.
25. Varshney R, Kale RK. Effects of calmodulin antagonists on radiation induced lipid peroxidation in microsomes. Int. J. Radiat. Biol. 1990;58(5):733-43.
26. Misra HP, Fridovich I. The role of superoxide anion in the autooxidation of epinephrine and a simple assay for superoxide dismutase. J. Biol. Chem. 1972;247:3170-3175.
27. Magwere T, Naik YS, Hasler TA. Effects of chloroquine treatment on antioxidants enzymes in rats liver and kidney. Free Radical. Biol. Med. 1997;22:321-327.
28. Sinha AK. Colorimetric assay of catalase. Anal. Biochem. 1972;47:389-394.
29. Habig WA, Pabst MJ, Jacoby WB. Glutathione transferases; the first enzymatic step in mercapturic acid formation. J. Biol. Chem. 1974;249:7130-7139.
30. Lowry OH, Rosebrough NJ, Farr AI, Randall RJ. Protein measurement with folin phenol reagent. J. Biol. Chem. 1951; 193:265-275.
31. Baker FJ, Silverton RE. Introduction to Medical Laboratory Technology 6th ed., London Butter Worth; 1985.
32. Dhamidharka VR, Nadeau CL, Cannon HW, Harris, Rosen S. Ciprofloxacin overdose: Acute renal failure with prominent apoptotic changes. Am J Kid Dis. 1998;31:710-712.
33. Clark DWJ, Deborah L, Lynda VW, Gillian LP, Saad AWS. Profiles of hepatic and dysrhythmic cardiovascular events following use of fluoroquinolone antibacterials: Experience from large cohorts from the drug safety research unit prescription monitoring database. Drug Safety. 2004;24:1143-1154.
34. Pouzauaud F, Dutot M, Martin C, Debray M, Warnet JM, Rat P. Age-dependent effects on redox status, oxidative stress, mitochondrial activity and toxicity induced by fluoroquinolones on primary cultures of rabbit tendon cells. Comp Biochem Physiol. Toxicol Pharmacol. 2006;143: 232-241.
35. Harris ED. Regulation of antioxidant enzymes. J Nutr. 1992;122:625-626.
36. Adaramoye OA, Osaimoje DO, Akinsanya AM, Nneji CM, Fafunso MA, Ademowo OG. Changes in antioxidant status and biochemical indices after acute administration of artemether, artemether-lumefantrine and halofantrine in rats. Nordic Pharmacological Society. Basic Clin. Pharmacol Toxicol. 2008;102:412–418.
37. Sivaraj A, Vinothkurmar P, Sathiyaraj K, Sundaresan S, Devi K, Senthikumar B. Hepato-protective potential of A*ndrographis paniculata* aqueous leaf extract on ethanol induced liver toxicity in albino rats. J. Appl. Pharm. Sci. 2011; 1(06):204-208.
38. Owoeye O, Adedara IA, Bakare OS, Adeyemo OA, Egun C, and Farombi EO. Kolaviron and vitamin E ameliorate hematotoxicity and oxidative stress in brains of prepubertal rats treated with an anticonvulsant phenytoin. Toxicol. Mechanisms and Meth. 2014;24(5):353-361.
39. Peltola V, Huhtaniemi I, Ahotupa M. Antioxidant enzyme activity in the maturing rat testis. J. Androl. 1992;13:450-455.
40. Masaki H, Okano Y, Sakurai H. Differential role of catalase and glutathione peroxidase in cultured human fibroblasts under exposure of H_2O_2 or ultraviolet B light. Arch. Dermatol. Res. 1998;290:113-118.
41. Trachootham D, Lu W, Ogasawara MA, Rivera-Del VN and Huang P. Redox Reg. of Cell Survival. Antioxid. Redox Signal. 2008;10(8):1343–1374.
42. Altınordulu S, Eraslan G. Effects of some quinolone antibiotics on malondialdehyde levels and catalase activity in chicks. Food Chem. Toxicol. 2009;47:2821–2823.
43. Rajurkar RB, Khan ZH, Gujar GT. Studies on levels of glutathione s-transferase, its isolation and purification from *Helicoverpa armingera*. Curr. Sci. 2003;85:1355-60.

44. Hayes JD and Pulford DJ. The glutathione S-transferase supergene family: Regulation of GST and the contribution of the isoenzymes to cancer chemoprotection and drug resistance. Crit. Rev. Biochem. Mol. Biol. 1995;30:445–600.
45. Marnett LJ. Generation of mutagens during arachidonic acid metabolism. Cancer Metastasis Reviews. 1994;13:303-308.
46. Sherratt PJ, Hayes JD. Glutathione S-transferases. Enzyme systems that metabolise drugs and other xenobiotics. Wiley and Sons, New York; 2001.
47. Jemec A, Lešer V, Drobne D. The link between antioxidant enzymes catalase and glutathione S-transferase and physiological condition of a control population of terrestrial isopod (*Porcellio scaber*). Ecotoxicol Environ Saf. 2012;79:42-7.
48. Vontas JG, Small GJ, Hemingway J. Glutathione S- transferase an antitoxicant defence agents confer pyrethroid resistance in *Nilaparavata lugens*. Biochem. J. 2001;357:65-72.
49. Adaramoye OA, Adeyemi EO. Hepatoprotection of d-galactosamine-induced toxicity in mice by purified fractions from *Garcinia kola* seeds. Basic Clin. Pharmacol. Toxicol. 2006;98:135-141.
50. Halliwell B. Reactive oxygen species and the central nervous system. J Neurochem. 1992;59(5):1609–23.
51. Masella R, Di Benedetto R, Vari R, Filesi C, Giovannini C. Novel mechanisms of natural antioxidant compounds in biological systems: Involvement of glutathione and glutathione-related enzymes. J Nutr. Biochem. 2005;16:577–586.
52. Curello S, Ceconi C, Bigoli C, Ferrari R, Albertini A, Guarnieri C. Changes in the cardiac glutathione status after ischemia and reperfusion. Experientia. 1985;41:42–43.
53. Lobo V, Patil A, Phatak A, Chandra N. Free radicals, Antioxidants and Functional Foods: Impact on Human health. Pharmacogn. Rev. 2010;4(8):118-126.
54. Suntres ZE. Liposomal Antioxidants for Protection against Oxidant-Induced Damage. J of Toxicol. 2011;152474:1-16.
55. Domitrović R. Vitamin C in disease prevention and therapy. Biochemia Medica. 2006;16(2):107-25.
56. Foyer CH, Noctor G. Ascorbate and Glutathione: The Heart of the Redox Hub. Plant Physiol. 2011;155(1):2-18.
57. Yagi K. Lipid peroxides and related radicals in clinical medicine. Adv Exp Med Biol.1994;366:1-15.
58. Pryor WA. Cigarette smoke radicals and role of free radicals in chemical carcinogenicity. Environ. Health Perspect. 1997;105(supp14):875-882.
59. Itoro FU, Itemobong SE, Oboso EE, Henry DA, Enomfon JA, Fakoya A. Antioxidant and hepatoprotective effects of dried flower extracts of *Hibiscus sabdariffa* L. on rats treated with carbon tetrachloride. J of Appl. Pharm. Sci. 2012;02(08):186-189.
60. Gutteridge JMC. Lipid peroxidation and antioxidants as biomarkers of tissue damage. Clin. Chem. 1995;41:1819–1828.
61. Uzma N, Kumar B, Salar K, Madhuri A, Reddy V. *In vitro* and *In vivo* evaluation of toxic effect of benzene on lymphocytes and hepatocytes. The Internet J of Toxicol. 2008;6(2).
62. Bigoniya P, Singh CS, Shukla A. Comprehensive review of different liver toxicants used in experimental pharmacology. International Journal of Pharm. Sci. Drug Res. 2009;1(3):124-135.
63. Mohan H. Text Book of Pathology, 4th ed; Jaypee Publications, New Delhi. 2000; 571.
64. Dey NC, Dey TK. A Text book of pathology, 15th ed; New Central Book Agency (P) Ltd; Calcutta. 2002;19.2.
65. Mohammed I, Khaja ZU and Mangamoori LN. Hepatoprotective activity of *Boswellia serrata extracts*: *In vitro* and *In vivo studies.* Inter. J of Pharm Applications. ISSN 0976-2639. 2011;2(Issue 1):89-98.
66. Bhattacharyya D, Mukherjee R, Pandit S, Das N, Sur TK. Prevention of carbonte trachloride induced hepatotoxicity in rats by Himoliv®, a polyherbal formulation. Ind. J. Pharmacol. 2003;35:183-185.
67. Friedman LS, Martin P, Muòoz SJ. Liver function tests and the objective evaluation of the patient with liver disease. In: Hepatology: A Textbook of Liver Disease. 3rd edn. Philadelphia: WB Saunders Co. 1996;1:791-833.
68. Tredger JM, Sherwood RA. The liver: New functional, prognostic and diagnostic tests. Annuals of Clinical Biochem. 1997;34 (Pt 2):121-41.

69. Khadabadi SS, Gond NY, Ghiware NB, Shendarkar GR. Hepatoprotective effects of *Ficus carica* leaf in chronic hepatitis. Indian Drugs. 2007;44:54-57.
70. Ritesh K, Nanda K, Swami KR, Srivastava ASV, Santosh KV, Srinath R. Hepatoprotective activity of alcoholic and aqueous extract of *Terminalia belerica* in Rats, Adv. Pharmacol. Toxicol. 2008;9:1-71.
71. Vahdati-Mashhadian NH, Rakhshandeh OA. An investigation on LD50 and subacute hepatic toxicity of Nigella sativa seed extracts in mice. Pharmazie. 2005; 60:544-547.
72. Ewaraiah MC, Satyanarayana T. Hepatoprotective activity of extracts from stem of *Mussaenda erythrophylla* Lam. Against carbon tetrachloride-induced toxicity in rats. JPRHC. 2010;2:23-31.
73. Gaspari F, Perico N, Matalone M, Signorini O, Azzollini N, Mister M, Remuzzi G. Precision of plasma clearance of iohexol for estimation of GFR in patients with renal disease. J. Am. Soc. Nephrol. 1998;9:310-313.
74. Nankivell BJ. Creatinine clearance and the assessment of renal function. Aust. Prescr. 2001;24:15-17.
75. Traynor J, Geddes CC, Fox JG. How to measure renal function in clinical practice. BMJ. 2006;333:733-737.
76. Perrone R, Madias N, Levy A. Serum Creatinine as an index of renal function: New insights into old concepts. Clin. Chem. 1992;38:1933-1953.
77. Mouton R, Holder K. Laboratory tests of renal function. Anaesthesia Intensive Care Med. 2006;7(7):240-243.

© 2015 Olayinka et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sciencedomain.org/review-history.php?iid=986&id=14&aid=8658