African Journal of Pharmacy and Pharmacology Vol. 5(16), pp. 1888-1897, 29 October, 2011
Available online at http://www.academicjournals.org/AJPP
DOI: 10.5897/AJPP11.417
ISSN 1996-0816 ©2011 Academic Journals

Full Length Research Paper

Effect of ciprofloxacin and levofloxacin on some oxidative stress parameters in brain regions of male albino rats

Sayed M. Rawi[1], Iman M. Mourad[2]*, Nadia M. S. Arafa[3] and Naema I. Alazabi[4]

[1]Faculty of Science and Arts, Khalius King Abdul Aziz University, Saudia Arabia.
[2]Department of Zoology, Faculty of science, Cairo University, Giza, Egypt.
[3]Faculty of Education S.D. (Girls), Jazan University, Saudia Arabia.
[4]Algharbe University, Gahrian, Libya.

Accepted 7 October, 2011

The present study aimed to investigate the possibility of involved oxidative stress due to the oral administration of either ciprofloxacin or levofloxacin (under therapeutic level) in the three brain regions, cerebral cortex, hippocampus and striatum of male albino rats weighing (100 ± 20 g). The ciprofloxacin and levofloxacin administered groups exhibited significant elevation in lipid peroxidation (LPO), nitric oxide (NO) contents and superoxide dismutase (SOD) enzyme activity in cortex, hippocampus and striatum. The redox status (reduced glutathione/oxidized glutathione) and glutathione peroxidase (GPx) and Na^+, K^+, -adenosine triphosphatase (Na^+, K^+, -ATPase) enzymes activity were significantly reduced in a dose dependant manner in the three brain regions. Generally, the data suggest the contribution between these antibiotics and oxidative stress in brain regions which through light on the need of studies for design and development of new quinolone derivatives with broader antibacterial activity and better pharmaco-kinetics avoiding central nervous system (CNS) side effects.

Key words: Ciprofloxacin, levofloxacin, oxidative stress, cerebral cortex, hippocampus, striatum.

INTRODUCTION

Fluoroquinolone antimicrobial agents are widely used for the treatment of various infections, because of their broad spectrum of antimicrobial activity (Owens and Ambrose, 2005), especially against Gram-negative bacteria, prescribed for respiratory and urinary tract infections (Oliphant and Green, 2002; Ball, 2003). They are generally regarded as safe drugs associated with mild gastrointestinal and central nervous system (CNS) symptoms (Bertino and Fish, 2000; Sprandel and Rodvold, 2003). They have been widely used in clinical practice for their excellent activities. They have been proposed as alternative in the treatment of CNS infections, because they can spread into the CNS (Hasbun and Quagliarello, 1998; Kim et al., 2009). In fact several clinical observations indicated the possible incidence of undesirable adverse reactions following quinolone use. Among the various adverse effects are those on CNS, including headache, confusion, hallucination, anxiety, nervousness, nightmares and convulsive seizures (Carrie, 2004; Saito et al., 2008).

Ciprofloxacin is a synthetic antibacterial agent belonging to the second generation of the family of fluoroquinolones, it well absorbed orally and it induced its antibacterial action mainly by inhibiting_DNA gyrase, a type II topoisomerase, and topoisomerase IV enzymes necessary to separate bacterial DNA, thereby inhibiting cell division (Akasaka et al., 1998; Liu and Wang, 1999).

Levofloxacin is a synthetic chemotherapeutic antibiotic of the fluoroquinolone drug class (Kawahara, 1998; Nelson et al., 2007). It is used for the treatment of infections of the respiratory and urinary tract, skin and soft tissues. Like other fluoroquinolones, it exerts antibacterial effect not exclusively in the blood but particularly in the inflamed tissue (Ungerstedt, 1991; Müller, 2000) and is used to treat severe or life-threatening bacterial infections or bacterial infections that

*Corresponding author. E-mail: yaramorad@ yahoo.com.

have failed to respond to other antibiotic classes (Liu and Mulholland, 2005; MacDougall et al., 2005).

Oxidative stress occurs because of an alteration in the equilibrium of the production of reactive oxygen species (ROS) and antioxidative processes in favor of the production of ROS. ROS include nonorganic molecules, such as the superoxide radical anion (O^{-2}), hydrogen peroxide (H_2O_2) and hydroxyl radicals (HO^-), as well as organic molecules, such as alkoxyl and peroxyl radicals. In order to avoid damage caused by ROS, like lipid peroxidation (LPO), protein modification and DNA strand breaks, mechanisms exist which remove ROS or prevent the generation of ROS (Sies, 1991; Halliwell, 1992). For example, the removal of superoxide and H_2O_2 prevents the generation of hydroxyl radicals, which formed by the iron-catalyzed Fenton reaction or by the Haber-Weiss-reaction (Winterbourn, 1995; Wardman and Candeias, 1996) and are the most reactive species within the ROS family.

Cell damages caused by free radicals appear to be major contributor to aging, cancer, cardiovascular disease, immune system decline, brain and liver dysfunction. All free radicals are highly reactive and highly damaging to biological system (Moein et al., 2007).

Oxidative stress generated by ROS appears connected with the loss of neurons during the progression of neurodegenerative diseases, that is, Parkinson's disease, Alzheimer's disease and amyotrophic lateral sclerosis (ALS) (Cadet and Brannock, 1998; Sun and Chen, 1998). These facts underline the importance of an effective antioxidant system for brain function during a long human life. Increase in oxidative stress was postulated to play an important role in the pathogenesis of a number of neurodegenerative diseases (Shelat et al., 2008).

Recently, a number of antibiotics, including ciprofloxacin, have been demonstrated to stimulate the production of ROS in bacterial cells (Becerra and Albesa, 2002; Albesa et al., 2004). Reactive oxygen species are reactive by-products formed by the partial reduction of molecular oxygen (Pomposiello and Demple, 2000). Redox cycling of various chemical substances, including some antibiotics, affects the ROS produced by cells during the oxidation process (Butler and Hoey, 1993). Fluoroquinolones are known to induce the formation of singlet oxygen ($^1O_2$) and superoxide anion (O_2^-), which are responsible for the phototoxic effect of the fluoroquinolones (Umezawa et al., 1997). In addition, the two prominent side effects of aminoglycoside antibiotics, ototoxicity and nephrotoxicity, are also believed to involve ROS (Brummett and Fox, 1989; Mingeot-Leclercq and Tulkens, 1999). A number of diverse cellular processes that lead to cell death are also mediated through ROS (Cabiscol et al., 2000).

The present study was designed to determine the possibility of oxidative stress due to the oral administration of either ciprofloxacin or levofloxacin (under therapeutic level) in brain regions of male albino rats, that is, through the determination of some of the antioxidant system parameters, lipid peroxidation (LPO), reduced glutathione (GSH), oxidized glutathione (GSSG), nitric oxide (NO) (nitrite/nitrate) in addition to superoxide dismutase (SOD), glutathione peroxidase (GPx) and sodium-potassium-adenosine triphosphatase (Na^+, K^+, -ATPase) enzyme activities and total protein in the three brain regions cerebral cortex, hippocampus and striatum.

MATRIALS AND METHODS

Experimental animals

This study was carried out on adult male albino rats, with average weight of range 100 ± 20 gm. They were obtained from the Egyptian Institution of Serum and Vaccine (Helwan). The experimental animals were allowed to acclimate under the laboratory conditions two weeks before the beginning of the experiments. The animals were kept under controlled temperature of 21°C and 12 h light/12 h dark cycle throughout the course of the experiment. A commercial pelleted diet and fresh vegetables were used before the experiment. The food debris, feces and urine were removed daily to prevent food and water contamination.

Drugs

The drugs used in the present work, were ciprofloxacin and levofloxacin, and they are as follows:

1. Ciprofloxacin (Cipro), manufactured by Bayer healthcare pharmaceuticals, ciprofloxacin hydrochloride. Tablets are synthetic broad spectrum antimicrobial agents for oral administration. Ciprofloxacin hydrochloride, United Stated Pharmacopoeia (USP), a fluoroquinolone, is the monohydrochloride monohydrate salt of 1-cyclopropyl-6-fluoro-1,4-dihydro-4-oxo-7-(1-piperazinyl)-3-quinoline carboxylic acid. It is a faintly yellowish to light yellow crystalline substance. The molecular weight is 385.8 and the empirical formula is $C_{17}H_{18}FN_3O_3 \cdot HCl \cdot H_2O$.
2. Levofloxacin, manufactured by Hoechst Marion Roussel Germany, The empirical formula is $C_{18}H_{20}FN_3O_4 \cdot ½\ H_2O$ and the molecular weight is 370.38. Levofloxacin is a light yellowish-white to yellow-white crystal or crystalline powder.

The antibiotic drugs were administered by gastric intubation technique and the administered doses were calculated equivalent to the human therapeutic dose according to the Food and Drug Administration (FDA) (Guidance for Industry and Reviewers, 2002).

Experimental design

The rats were divided into three main groups, each contain eight rats. The rats of the first group were administered 2 ml of distilled water daily (control), whereas those of the second group were administered daily 80 mg/kg body weight ciprofloxacin dissolved in 2 ml of water (Cipro-treated rats). The rats of the third group were daily administered 40 mg/kg body weight levofloxacin dissolved in 2 ml water (Levo-treated rats).

At the end of the experimental periods 3, 7 and 14 days animals were sacrificed after 12 h from the last dose by rapid decapitation and brain areas, the cerebral cortex, hippocampus and striatum were separated from each whole brain. Each brain area was divided into two halves the first half served for the enzymes activity assays and total protein. Superoxide dismutase (SOD) was determined according to Minami and Yoshikawa (1979) method.

Table 1. Effect of oral administration of ciprofloxacin (80mg/kg) and levofloxacin (40mg/kg) on some oxidative stress parameters in the cortex of male albino rats.

Parameter	Treated group	Time period (days)		
		3	7	14
MDA(nmol/g tissue)	Control	106.57 ± 3.13	107.60 ± 2.96	108.60 ±2.36
	Ciprofloxacin	150.15 ± 3.70*	144.67 ± 2.33*	138.33 ± 4.04*a
	levofloxacin	147.34 ± 3.02*	134.32±4.11*a	150.25 ± 4.49*b
(GSH/GSSG) (nmol/g)	Control	4.40 ± 0.41	4.45 ± 0.29	4.27 ± 0.33
	Ciprofloxacin	3.47 ± 0.34*	3.13 ± 0.15*	2.81 ± 0.29*ab
	levofloxacin	3.21 ± 0.29*	2.80± 0.24*a	2.38 ± 0.15*ab
NO (mg/g tissue)	Control	0.83 ± 0.08	0.83 ± 0.06	0.83 ± 0.08
	Ciprofloxacin	2.07 ± 0.04*	1.45 ± 0.19*a	1.50 ± 0.23*a
	levofloxacin	1.42 ± 0.07*	1.56± 0.24*	1.40 ± 0.11*b
SOD (U/mg protein)	Control	0.72 ± 0.06	0.72 ± 0.06	0.725 ± 0.06
	Ciprofloxacin	0.78 ± 0.06	1.11 ± 0.10*a	1.22 ± 0.10*ab
	levofloxacin	0.84 ± 0.12*	1.28±0.11*a	1.32 ± 0.10*a
GPx (mmol of GSH oxidized/min/mg protein	Control	1.32 ± 0.10	1.34 ± 0.07	1. 34 ± 0.06
	Ciprofloxacin	1.25 ± 0.12	0.99 ± 0.07*a	0.91 ± 0.04*ab
	levofloxacin	1.16 ± 0.06*	0.89 ± 0.03*a	0.75 ± 0.08*ab
Na^+, K^+, -ATPase (µmol Pi/mg protein)	Control	6.29 ± 0.32	6.25 ± 0.25	6.21 ± 0.34
	Ciprofloxacin	5.91 ± 0.33*	5.62 ± 0.35*	4.56 ± 0.34*ab
	levofloxacin	6.01 ± 0.48	5.07 ± 0.34*a	4.39 ± 0.22*ab

Data are expressed as Mean ± SE. Number of animals in each group is 8. P > 0.05 non-significant, *P < 0.05 significant, the symbol represent statistical significance; (a) significant as compared to 3 days treatment, (b) Significant as compared to 7days.

According to modified procedure of Rotruck et al. (1973) glutathione peroxidase (GPx) activity was determined. The measurement of (Na^+, K^+- ATPase) activity was determined according to the method described (Atkinson et al., 1971; Tsakiris, 2001). The method of Lowry et al. (1951) was used in the estimation of protein content of tissue homogenates.

The second half of each brain area was homogenized in 75% high performance liquid chromatography (HPLC) methanol (1/10 weight/volume) using a homogenizer surrounded with an ice jacket and the homogenates were used for the determination of the brain areas contents of malondialdehyde (MDA) as index for lipid peroxidation (LPO), glutathione reduced (GSH), glutathione oxidized (GSSG) and nitric oxide (NO) as (NO_2/NO_3) were determined by HPLC (Karalas et al., 2002; Jayatilleke and Shaw, 1993; Papadoyannis et al., 1999), respectively. The assays used Agilent thermostated 1200 series HPLC system from Agilent Technologies (USA) with Agilent chemstation chromatography data system software.

Statistical analysis

The data were expressed as means ± standard error of the mean (SEM). All variables were tested for normal distribution using t test (P < 0.05) and one way analysis of variance (ANOVA). Statistical analysis was evaluated according to Duncan's test to clarify the experimental periods' groups. Statistical Processor System Support "SPSS" for Windows software, Release 17.0 (SPSS, Chicago, IL) was used.

RESULTS

The present study showed the effect of oral administration of 80 mg/kg body weight of ciprofloxacin and 40 mg/kg body weight of levofloxacin in the cortex, hippocampus and striatum of male albino rats at different three time periods.

The data represented that LPO level increased significantly throughout the experimental periods of 3, 7 and 14 days after the oral administration of ciprofloxacin and levofloxacin in the cortex, hippocampus and striatum tissues as compared to the control values.

The recorded data in Tables 1 and 2 revealed significant decrease in the redox status (GSH/GSSG) in the cortex and the hippocampus of male albino rats for both treatment after the 3rd , 7th and 14th days, respectively, as compared to the control values. On the other hand, in the striatum the data showed significant (P < 0.05) decrease in the ratio after the administration of

Table 2. Effect of oral administration of ciprofloxacin (80 mg/kg) and levofloxacin (40 mg/kg) on some oxidative stress parameters in the hippocampus of male albino rats.

parameters	Treated groups	Time period (days)		
		3	7	14
MDA(nmol/g tissue)	Control	93.58 ± 1.99	96.08 ± 1.82	94.71 ± 1.87
	Ciprofloxacin	146.91 ± 2.56*	155.43 ±2.07*	157.09 ± 3.42*[a]
	levofloxacin	141.01 ± 3.20*	153.49±3.85*[a]	152.20 ±3.36*
(GSH/GSSG) (❍mol/g)	Control	4.14 ± 0.33	4.17 ± 0.38	4.09± 0.38
	Ciprofloxacin	3.67 ± 0.29*	3.37 ± 0.30*	3.10 ± 0.33*[a]
	levofloxacin	3.44 ± 0.33*	3.05± 0.26*	2.85± 0.27*[a]
NO (mg/g tissue)	Control	0.44 ± 0.02	0.46 ± 0.03	0.44 ± 0.02
	Ciprofloxacin	0.72 ± 0.06*	0.84 ± 0.09*[a]	1.27 ± 0.07*[ab]
	levofloxacin	1.28 ± 0.09*	1.31±0.08*	1.25 ± 0.09*
`SOD (U/mg protein)	Control	1.67 ± 0.14	1.67 ± 0.13	1.63± 0.13
	Ciprofloxacin	1.65 ± 0.08	1.82 ± 0.09*[a]	1.73 ± 0.11
	levofloxacin	1.67 ± 0.16	1.67 ±0.13	1.83 ± 0.13*[ab]
GPx (mmol of GSH oxidized/min/mg protein	Control	1.36 ± 0.11	1.34 ± 0.11	1.34 ± 0.09
	Ciprofloxacin	1. 25 ± 0.09*	1.12 ± 0.10*[a]	1.05 ± 0.05*[a]
	levofloxacin	1.20 ± 0.12*	1.09±0.06*[a]	0.87 ± 0.05*[ab]
Na^+, K^+, -ATPase) (µmol Pi/mg protein)	Control	8.12 ± 0.56	8.09± 0.42	7.95 ± 0.46
	Ciprofloxacin	7.84 ± 0.46	7.75 ± 0.47	6.68 ± 0.27*[ab]
	levofloxacin	7.26 ± 0.72*	6.99±0.58*	6.69 ± 0.47*[a]

Data expressed as M ± SE. Number of animals in each group is 8. P > 0.05 non-significant, *P < 0.05 significant. The symbol represents statistical significance; (a) significant as compared to 3 days treatment, (b) significant as compared to 7days.

ciprofloxacin only after 14 days of treatment, while levofloxacin administration caused a significant decrease in (GSH/GSSG) from the 3rd day till the 14th days.

As regard to the effect of ciprofloxacin and levofloxacin on NO contents of male albino rats, the data showed significant increase from the 3rd day till the end of the experimental period in three brain areas after ciprofloxacin and levofloxacin administration as compared to the corresponding control levels.

The general pattern about the data of SOD enzyme activity in the cortex, hippocampus and striatum tissues of male albino rats affected by ciprofloxacin and levofloxacin administration are given in Tables 1 to 3. The data showed significant increase (P < 0.05) in SOD activity in the cortical and striatal tissues from the 3rd day achieving the maximum activity in the 14th day of ciprofloxacin treatment as compared to the control enzyme activity. Levofloxacin administration behaved like ciprofloxacin administration by the significant increase in the SOD activity level from the 3rd day till the 14th day as compared to the corresponding control enzyme activities. In the hippocampus tissue, the data showed significant increase in SOD activity after the 7 and 14th days of ciprofloxacin administration as compared to the control. In the same trend, levofloxacin administration exhibited significant increase at 0.05 levels in SOD activity from the7th duration value to the 14th day's groups as compared to control value.

Regarding to GPx enzyme activity, the data in Table 1 showed significant decrease (P < 0.05) in GPx activity in the cortical tissue from the 7th day achieving the minimum activity in the 14th day of ciprofloxacin treatment as compared to the control values. Levofloxacin administration behaved like ciprofloxacin administration by the significant decrease in the GPx activity from the 3rd day till the 14th day as compared to the corresponding control. In the hippocampus tissue, the data showed significant decrease in the GPx activity in the 3rd, 7th and 14th days of both treatments as compared to the corresponding control enzyme activity values.

On the other hand, the data in Table 3 showed significant decrease (P < 0.05) in GPx activity in the striatum only after the 14th day of ciprofloxacin administration and after 7 and 14th days of levofloxacin

Table 3. Effect of oral administration of ciprofloxacin (80 mg/kg) and levofloxacin (40 mg/kg) on some oxidative stress parameters in the striatum of male albino rats.

parameter	Treated group	Time period (days)		
		3	7	14
MDA (nmol/g tissue)	Control	128.11 ± 1.67	127.43 ± 1.70	128.11 ±1.15
	Ciprofloxacin	227.19 ± 4.24*	213.32 ± 5.14*a	236.36 ± 5.76*b
	levofloxacin	243.04 ± 2.81*	214.73 ± 5.99*a	218.19 ± 4.39*a
(GSH/GSSG) (nmol/ g)	Control	2.41 ± 0.19	2.27 ± 0.25	2.40 ± 0.19
	Ciprofloxacin	2.31 ± 0.15	2.27 ± 0.18	2.15 ± 0.15*
	levofloxacin	2.28 ± 0.04*	2.15 ± 0.08*	1.96 ± 0.08*ab
NO (mg/g tissue)	Control	0.81 ± 0.06	0.78 ± 0.07	0.82 ± 0.07
	Ciprofloxacin	1.16 ± 0.12*	0.95 ± 0.07*a	1.08 ± 0.07*b
	levofloxacin	1.12 ± 0.09*	1.35 ± 0.13*a	1.15 ± 0.08*b
SOD (U/mg protein)	Control	1.30 ± 0.07	1.28 ± 0.06	1.28 ± 0.05
	Ciprofloxacin	1.52 ± 0.08*	1.70 ± 0.10*a	1.66 ± 0.07*a
	levofloxacin	1.56 ± 0.11*	1.69 ± 0.09*a	1.84 ± 0.15*ab
GPx (mmol of GSH oxidized/min/mg protein	Control	2.41 ± 0.14	2.32 ± 0.13	2.42 ± 0.13
	Ciprofloxacin	2.29 ± 0.22	2.24 ± 0.10	2.14 ± 0.17*
	levofloxacin	2.28 ± 0.10	2.13 ± 0.24*	2.06 ± 0.16*a
Na^+, K^+, -ATPase) (µmol Pi/mg protein)	Control	6.91 ± 0.43	6.89 ± 0.24	6.81 ± 0.41
	Ciprofloxacin	6.64 ± 0.61	6.38 ± 0.61*	6.35 ± 0.50
	levofloxacin	6.90 ± 0.60	6.80 ± 0.37	6.45 ± 0.27

Data expressed as M ± SE. Number of animals in each group is 8. P > 0.05 non-significant, *P < 0.05 significant, the symbol represent statistical significance; (a) significant as compared to 3 days treatment, (b) significant as compared to 7days.

administration compared to the control values. Table 1 showed that the ATPase enzyme activity decreased in the cortical tissue with significant decrease ($P < 0.05$) from the 3rd day achieving the minimum activity in the 14th day of ciprofloxacin treatment as compared to the control. Levofloxacin administration showed significant decrease in the ATPase activity in the 7th and 14th days as compared to the corresponding control values.

In the hippocampus tissue, the data showed significant decrease in the ATPase activity only after the 14th day of ciprofloxacin administration as compared to the control enzyme activity. On the other hand, levofloxacin administration exhibited significant decrease at 0.05 levels in ATPase activity from the 3rd duration value to the 14th one as compared to the corresponding control values.

However, the striatum data showed no significant effect in the ATPase activity throughout the experimental duration period, in which the administration of either ciprofloxacin or levofloxacin was compared to the control enzyme activities except only the significant decrease at the 7th day of ciprofloxacin administration as compared to the corresponding control value.

DISCUSSION

Fluoroquinolones are among the most widely prescribed antibiotics, especially for respiratory and urinary tract infections. They are generally regarded as safe drugs associated with mild gastrointestinal and CNS symptoms (Jose et al., 2007; Becnel et al., 2009).

Ciprofloxacin remains amongst the safest of all antibiotics with remarkably few reports of serious reactions over a period of 15 years of use and more than 340 million prescriptions (Ball et al., 1999; Segev et al., 1999). Ciprofloxacin-associated seizures occur most commonly in patients with special risk factors that may cause accumulation of drug (high doses of the drug, old age, renal insufficiency, drug interactions) or that may decrease the threshold of epileptogenic activity (Agbaht et al., 2009). Levofloxacin is the *l*(-) isomer of the fluoroquinolone ofloxacin. Ofloxacin is a racemic mixture

of two optical isomers, *d*(+) and *l*(-) ofloxacin, which exist in a ratio of approximately 1:1. Of the four two isomers, only the *l*(-) form, levofloxacin, exhibits antibacterial activity.

Quinolones are known to induce cell death through the introduction of double-stranded DNA breaks following arrest of topoisomerase function (Drlica et al*.,* 2008). The bactericidal action of quinolones promote the generation of lethal hydroxyl radicals in both Gram-negative and Gram-positive bacteria, despite the stark differences in their primary drug- target interactions (Kohanski et al*.,* 2007). Studies suggested that antibiotic induced hydroxyl radical formation is the end product of a common mechanism, in which alterations in central metabolism related to NADH, nicotinamide adenine dinucleotide (NADH) consumption (increased TCA cycle and respiratory activity) are crucial to superoxide-mediated iron-sulphur cluster destabilization and stimulation of the Fenton reaction (Kohanski et al., 2010).

Brain has several characteristics that make it susceptible to free radical mediated injury. Brain lipid are highly enriched in polyunsaturated fatty acids, additionally, brain is critically dependent on aerobic metabolism and the mitochondrial respiratory activity is higher than that in many other tissues, increasing the risk of free radical "leak" from the mitochondria. The brain processes large amounts of O_2 in relatively small mass, and has a high content of substrates available for oxidation in conjunction with low antioxidant activities, making it extremely susceptible to oxidative damage (Bergamini et al., 2004). In addition, certain regions of central nervous system (CNS), such as the hippocampus, may be particularly sensitive to oxidative stress, because of their low endogenous levels of vitamin E, an important biochemical antioxidant, relatively to other brain regions (Henderson et al*.,* 1999; Gottlieb et al*.,* 2006). Such a depressed defense system may be adequate under normal circumstances. However, under pro-oxidative conditions, like seizures, these low antioxidant defenses can predispose the brain to oxidative stress.

The present study aims to throw light on the possibility of involved oxidative stress effect of either ciprofloxacin or levofloxacin, under therapeutic level, in three brain regions of male albino rat. The study extended to the three brain areas as the cortex, and hippocampus areas appeared to be important in the expression of early convulsive seizures (Kelly et al*.,* 1999; Ang et al*.,* 2006) in addition to the important functional association between cortical regions and the hippocampus in seizure propagation (Cavalheiro et al*.,* 1991; Kelly et al., 2002). The cortex and hippocampus suggested playing a role in inducing convulsions by quinolones (Motomura et al*.,* 1991) and there are direct anatomical connections between the hippocampus and the striatum (Voorn et al., 2004). In addition, there are connections between striatum and hippocampus via the entorhinal and prefrontal cortex (Hyman et al., 1990; Christakou et al., 2004).

Lipid peroxidation in a tissue is an index of irreversible biological damage of the cell membrane phospholipid, which in turn leads to inhibition of most of the sulphydryl and some nonsulphydryl enzymes. There is a close relationship that exists between inhibition of ATPase enzyme activities and elevation of LPO (Sawas and Gilbert, 1984; Hayashi, 2009). Our study recorded that LPO marker (MDA) levels in the cortex, hippocampus and striatum of adult male rats were increased significantly following administration of either ciprofloxacin or levofloxacin. This was done along with the significant decrease in ATPase enzyme activity in a dose dependent manner in cortex and hippocampus after either ciprofloxacin or levofloxacin administration and in striatum only after ciprofloxacin administration. The redox status (GSH/GSSG) and GPX enzyme activity reduced significantly, while SOD enzyme activity and NO level increased significantly in cortex, hippocampus and striatum after either ciprofloxacin or levofloxacin administration.

The increased MDA and NO levels and SOD enzyme activity in addition to the reduced level of glutathione demonstrated in this study in brain areas of either ciprofloxacine or levofloxacin treated animals along the lines of that recorded in different brain areas of the rat model of seizure induction (Munhoz et al., 2005; Rajasekaran, 2005; Gupta et al., 2009). The possible oxidative stress inducing effect of a fluoroquinolone antibiotic, ciprofloxacin was investigated in rats measuring glutathione redox status. Ciprofloxacin induced dose-dependent alterations in the glutathione redox status in liver and brain tissues (Gürbay and Hincal, 2004). Also, it has been shown that seizures cause an increase in the production of nitrite, a potent free radical known to be cytotoxic to neurons and glial cells (Freitas et al., 2005) which predicted in the brain areas results of this study supporting the oxidative stress inducing effects of these antibiotics (Gürbay et al., 2007; Li et al.; 2010) which may be concomitant with the previously discussed seizure associated effect of ciprofloxacin and levofloxacin (Darwish, 2008; Bellon et al*.,* 2009) and may add to reaffirming the hypothesis of LPO produced during seizure activity postulated for the fluoroquinolones. This may be explained via the previously mentioned mechanism of its bactericidal activity since ROS have a role in the antibacterial action of fluoroquinolones. As such, the effect of GSH/GSSG confirmed by the glutathione-mediated protection is specific to fluoroquinolones (Goswami et al., 2006). Cao et al. (2007) discussed the protective role of the glutathione content through the interpretation between the dopaminergic neuron injury and neuronal oxidative stress. Ciprofloxacin antimicrobial activity is based on the inhibition of bacterial DNA gyrase. However, this drug has also been found to affect mammalian topoisomerase II, especially its mitochondrial isoform. This impairs

mitochondrial DNA (mtDNA) handling and eventually results in a gradual decrease in mtDNA content in ciprofloxacin-treated cells (Castora et al., 1983; Lawrence et al., 1993). As mtDNA encodes 13 proteins, all engaged in oxidative phosphorylation, it is obvious that aberrant expression of mitochondrial genes and/or partial depletion of mtDNA may affect mitochondrial energy metabolism. In fact, disturbance of mitochondrial respiration and ATP synthesis has been postulated to explain the cytotoxic effect of ciprofloxacin which is in a concentration-dependent manner (Koziel et al., 2006). When ciprofloxacin is applied, concentrations that are insufficient for induction of apoptosis may stop cell proliferation by inhibition of mitosis. Chromosomal instability of such cells may, at least potentially, increase a risk of cancer development (Koziel et al., 2010).

The structural similarities of the fluoroquinolones to kynurenic acid and other similar compounds, which are endogenous ligands of the glutamate receptor, might suggest an interaction of quinolones with ligand-gated glutamate receptors as well (Schmuck et al .,1998) which may explain the excitatory effects on quinolones subjected groups. It has been shown that fluoroquinolones decrease blocking effects of Mg^{2+} and MK-801 binding to the N-methyl-D-aspartate (NMDA) type of glutamate receptors. Magnesium chelating properties of fluoroquinolones have been postulated as mechanisms of fluoroquinolone-induced atrophy, and the excitatory potency of fluoroquinolones might also be based on activation of the (NMDA) receptor by abolishing the Mg^{2+} block in the ion channel. This would prolong the opening time of the channel, thus increasing intracellular Ca^{2+} concentration in the neurons (Sen et al., 2007). The contribution of the release of excitatory amino acids may also be responsible for the large calcium-dependent increase in extra cellular potassium seen after experimental brain injury. Increased extra cellular potassium further increases neuronal excitability and may contribute to epileptogenesis (Nilson et al., 1994). The decrease in hippocampal ATPase activity throughout the experimental periods, post the antibiotics administration in line with the assumption of Machado et al. (2011) about the reduction of glutamate uptake and Na^+, K^+-ATPase activity in rat hippocampus may be mediated via oxidative stress.

Another explanation for the fluoroquinolones under investigation related oxidative stress effects via disorders of glucose homeostasis which have been reported in association with fluoroquinolone therapy (Tomita et al., 2007; Garber et al., 2009). Glucose homeostasis is significantly greater with gatifloxacin and levofloxacin, but not ciprofloxacin (Aspinall et al., 2009). Maeda et al. (1996) concluded that quinolone antibiotics may cause hypoglycaemia by increasing insulin release via blockade of K^+-ATP channels. Severe hypoglycaemia may cause neurological disturbances (Lawrence et al., 2006). During hypoglycemia, the excitatory and inhibitory nerve terminals contacting these neurons are enriched with aspartate, which may be released to activate NMDA receptors on synaptic and extrasynaptic sites. This would cause an excitotoxic insult, leading to neuronal death (Wieloch, 1985). The difference in the glucose homeostasis effect between levofloxacin and ciprofloxacine may through light as a cause for the significant effects between the recorded antioxidants results in levofloxacin treated groups over that of ciprofloxacin treated ones (Vallurupalli et al., 2008). The other expected cause may be related to the suggestion that levofloxacin crosses the blood-brain and/or blood-cerebrospinal fluid barriers more easily than ciprofloxacine does (Akahane et al., 1993, 1996).

Glucose metabolism via the pentose cycle plays a crucial role in providing nicotinamide adenine dinucleotide phosphate (NADPH) and, hence, maintaining the normal ratio of GSH to GSSG and a normal (GSH/GSSG) in cells. When the intracellular concentration of GSH decreases and that of GSSG increases, the cellular demand for NADPH increases markedly. This necessitates an increase in glucose metabolism via the pentose cycle. A deficiency of intracellular NADPH may exacerbate an imbalance between the production and scavenging of free radicals. When rates of free radical production are greater than the scavenging rates, oxidative damage likely occurs in cells and tissues (Sies, 1999).

Studies have shown that changes in the intracellular milieu of the cells, such as alterations in the redox environment, are important regulators of the progression to apoptosis (Pervaiz and Clement, 2002). Glutathione (GSH) depletion is an early hallmark in the progression of cell death in response to a variety of apoptotic stimuli in numerous cell types (Franco et al., 2007; Circu and Aw, 2008).

Glutathione content has been shown to regulate the activation of ionic conductances during apoptosis. Alterations in the intracellular ionic homeostasis of the cell have been clearly linked to the progression of apoptosis (Franco et al., 2006). Recent reports show that during apoptosis, cell shrinkage or apoptotic volume decrease, associated with the impairment in the Na^+, K^+-ATPase activity (Yin et al., 2007), and activation of K^+ and Cl^- conductances are also regulated by alterations in GSH homeostasis (Shimizu et al., 2004; Franco et al., 2007). A wide variety of ion channels and transporters have been shown to be regulated by ROS (Kourie, 1998). In the nervous system, GSH is necessary for the defense against oxidative stress and the progression of distinct pathologies including those of Parkinson's disease (PD) and Alzheimer's disease (AD). Alterations in intracellular GSH homeostasis have been observed in different experimental models of neurodegenerative diseases and a direct link between a reduction in GSH content and apoptosis in neuronal cells associated with neurodegeneration (Dringen and Hirrlinger, 2003; Diaz-

Hernandez et al., 2005).

Ciprofloxacin induced dose-dependent alterations in GSH/GSSG in the investigated brain areas in line with the report of Gürbay and Hincal (2004), and also the ciprofloxacin concentration-dependent induction of DNA damage in astrocytes (Gürbay et al., 2006).

Conclusion

Conclusively, a contribution between fluoroquinolones antibiotics and oxidative stress is not surprising. A tactic to enhance antibiotic efficacy by parallel treatments that compromise the oxidative effect must need intense investigation which may be by the use of antioxidants in line with the antibiotic treatments, as well as tactic through studies for design and development of new quinolone derivatives with broader antibacterial activity and better pharmaco-kinetics avoiding CNS side effects.

REFERENCES

Agbaht K, Bitik B, Piskinpasa S, Bayraktar M, Topeli A (2009). Ciprofloxacin-associated seizures in a patient with underlying thyrotoxicosis: case report and literature review. Int. Clin. Pharmacol. Ther., 47 (5): 303-310.

Akahane K, Kato M, Takayama S (1993). Involvement of Inhibitory and Excitatory Neurotransmitters in Levofloxacin- and Ciprofloxacin-Induced Convulsions in Mice. Antimicrob. Agents Chemother. 37(9): 1764-1770.

Akahane K, Ohkawara S, Nomura M, Kato M. (1996). Effect of bile duct ligation and unilateral nephrectomy on brain concentration and convulsant potential of the quinolone antibacterial agent levofloxacin in rats. Fundam. Appl. Toxico., 29: 280-286.

Akasaka T, Kurosaka S, Uchida Y, Tanaka M, Sato K, Hayakawa I (1998). Antibacterial activities and inhibitory effects of sitafloxacin (DU-6859a) and its optical isomers against type II topoisomerases. Antimicrob. Agents Chemother. 42: 1284-1287.

Albesa I, Becerra MC, Battan PC, Paez PL (2004). Oxidative stress involved in the antibacterial action of different antibiotics. Biochem. Biophys. Res. Commun. 317**:** 605-609.

Ang CW, Carlson GC, Coulter DA (2006). Massive and specific dysregulation of direct cortical input to the hippocampus in temporal lope epilepsy. J. Neurosci., 15, 26 (46): 11850-11856.

Aspinall SL, Good CB, Jiang R, McCarren M, Dong D, Cunningham FE (2009). Severe dysglycemia with the fluoroquinolones: a class effect? Clin. Infect. Dis. 49(3): 402-408.

Atkinson A, Gatenby AD, Lowe AG (1971). Subunit structure of Na+,K+ -dependent transport AT Pase. Nat. New Biol. 233: 145-146.

Ball P (2003). Adverse drug reactions: implications for the development of fluoroquinolones. J. Antimicrob. Chemother., 51: 21-27.

Ball P, Mandell L, Niki Y, Tillotson G (1999). Comparative tolerability of the newer fluoroquinolone antibacterials. Drug Saf., 21: 407-421.

Becerra MC, Albesa I (2002). Oxidative stress induced by ciprofloxacin in *Staphylococcus aureus*. Biochem. Biophys. Res. Commun., 297: 1003-1007.

Becnel BL, Maynard MJ, Morgan-Linnell SK, Horton LB, Sucgang R, Hamill RJ, Jimenez JR, Versalovic J, Steffen D, Zechiedrich L (2009). Relationships among ciprofloxacin, gatifloxacin, levofloxacin, and norfloxacin MICs for fluoroquinolone-resistant *Escherichia coli* clinical isolates. Antimicrob. Agents Chemother., 53(1): 229-234.

Bellon A, Perez-Garcia G, Coverdale JH, Chacko RC (2009). Seizures associated with levofloxacin: case presentation and literature review. Eur. J. Clin. Pharmacol., 65 (10): 959-962.

Bergamini CM, Gambetti S, Dondi A, Cervellati C (2004). Oxygen, reactive oxygen species and tissue damage, Curr. Pharm . Des., 10(14): 1611-1626.

Bertino JJR, Fish D (2000). The safety profile of the fluoroquinolones. Clin. Ther., 22: 798-817.

Brummett R, Fox K (1989). Aminoglycoside-induced hearing loss in humans. Antimicrob. Agents Chemother., 33: 797-800.

Butler J, Hoey BM (1993). Redox cycling drugs and DNA damage. In : Halliwell B. Aruoma OI (eds) DNA and free radicals: Ellis Horwood Ltd., West Sussex, England, pp. 243-273.

Cabiscol E, Tamarit J, Ros J (2000). Oxidative stress in bacteria and protein damage by reactive oxygen species. Int. Microbiol., 3: 3-8.

Cadet JL, Brannock C (1998). Free radicals and the pathobiology of brain dopamine systems. Neurochem. Int., 32: 117-131.

Cao Q, Wei LR, Lu LL, Zhao CL, Zhao HY, Yang H (2007). Astrocytes protect MN9D neuronal cells against rotenone-induced oxidative stress by a glutathione-dependent mechanism. Sheng Li Xue Bao., 59 (3): 253-259.

Carrie AQ (2004). Possible Gatifloxacin-Induced Seizure. Ann. Pharmacother. 38 (2): 235-237.

Castora FJ, Vissering FF, Simpson MV (1983). The effects of bacterial DNA gyrase inhibitors on DNA synthesis in mammalian mitochondria. Biochim. Biophys. Acta, 740: 417-427.

Cavalheiro EA, Leite JP, Bortolotto ZA, Turski WA, Ikonomidou C, Turski L (1991). Long-term effects of pilocarpine in rats: structural damage of the brain triggers kindling and spontaneously recurrent seizures. Epilepsia, 32: 778-782.

Christakou A, Robbins TW, Everitt B (2004). Prefrontal cortical-ventral striatal interactions involved in affective modulation of attentional performance: implications for corticostriatal circuit function. J. Neurosci., 24: 773-780.

Circu ML, Aw TY (2008). Glutathione and apoptosis. Free Radic. Res., 42: 689-706.

Darwish T (2008). Ciprofloxacin-induced seizures in a healthy patient. N Z Med. J., 121 (1277):104-105.

Diaz-Hernandez JI, Almeida A, Delgado-Esteban M, Fernandez E, Bolanos JP (2005). Knockdown of glutamate-cysteine ligase by small hairpin RNA reveals that both catalytic and modulatory subunits are essential for the survival of primary neurons. J. Biol. Chem., 280: 38992–39001.

Dringen R, Hirrlinger J (2003). Glutathione pathways in the brain. Biol. Chem., 384: 505-516.

Drlica K, Malik M, Kerns RJ, Zhao X (2008). Quinolone- mediated bacterial death. Antimicrob. Agents Chemother., 52: 385-392.

Franco R, Bortner CD, Cidlowski JA (2006). Potential roles of electrogenic ion transport and plasma membrane depolarization in apoptosis. J. Membr. Biol., 209: 43-58.

Franco R, DeHaven WI, Sifre M, Bortner CD, Cidlowski JA (2007). Glutathione depletion and disruption of intracellular ionic homeostasis regulate lymphoid cell apoptosis. J. Biol. Chem., 283: 36071-36087.

Freitas RM, Vasconcelos SM, Souza FC, Viana GS, Fonteles MM (2005). Oxidative stress in the hippocampus after status epilepticus in rats. FEBS J. 272: 1307-1312.

Garber SM, Pound MW, Miller SM (2009). Hypoglycemia associated with the use of levofloxacin. Am. J. Health Syst. Pharm. 66 (11): 1014-1019.

Goswami M, Mangoli SH, Jawali N (2006). Involvement of reactive oxygen species in the action of ciprofloxacin against *Escherichia coli*. Antimicrob. Agents . Chemother., 50 (3): 949-954.

Gottlieb M, Leal-Campanario R, Campos-Esparza MR, Sánchez-Gómez MV, Alberdi E, Arranz A, Delgado-García JM, Gruart A, Matute C (2006). Neuroprotection by two polyphenols following excitotoxicity and experimental ischemia. Neurobiol Dis., 23(2): 374-386.

Guidance for Industry and Reviewers (2002). Estimating the safe starting dose in clinical trials for therapeutics in adult healthy volunteers. Food and Drug Administration, pp.1-26.

Gupta YK, Briyal S, Sharma M (2009). Protective effect of curcumin against kainic acid induced seizures and oxidative stress in rats. Ind. J. Physiol. Pharmacol., 53(1): 39-46.

Gürbay A, Hincal F (2004). Ciprofloxacin-induced glutathione redox status alterations in rat tissues. Drug Chem. Toxicol., 27(3): 233-242.

Gürbay A, Gonthier B, Signorini-Allibe N, Barret L, Favier A, Hincal F (2006).Ciprofloxacin-induced DNA damage in primary culture of rat astrocytes and protection by Vitamin E. Neurotoxicology, 27(1): 6-10.

Gürbay A, Gonthier B, Barret L, Favier A, Hincal F (2007). Cytotoxic effect of ciprofloxacin in primary culture of rat astrocytes and protection by Vitamin E. Toxicology, 229 (1-2): 54-61.
Halliwell B (1992). Reactive oxygen species and the central nervous system. J. Neurochem., 59:1609-1623.
Hasbun R, Quagliarello VI (1998). Use of quinolones in the treatment of bacterial meningitis, In The quinolones 2nd ed. Academic Press, Inc., New York, N.Y, pp 2877-301.
Hayashi M (2009). Oxidative stress in developmental brain disorders. Neuropathol., 29: 1-8.
Henderson GI, Chen JJ, Schenker S (1999). Ethanol, oxidative stress, reactive aldehydes, and the fetus. Frontiers in Bioscience: J. Virt. Lib., 4: 541-550.
Hyman BT, Van Hoesen GW, Damasio A (1990). Memory-related neural systems in Alzheimer's disease: an anatomic study. Neurology, 40: 1721-1730.
Jayatilleke E, Shaw S (1993). A high performance liquid chromatographic assay for reduced and oxidized glutathione in biological samples. Anal. Biochem., 214 (2): 452-457.
Jose J, Jimmy B, Saravu K (2007). Dysglycemia associated with the use of fluoroquinolones-focus on gatifloxacin. J. Clin. Diag. Res., (3): 185-187.
karalas F, Karatepe M, Baysar A (2002). Determination of free malondialdehyde in human serum by high performance liquid chromatography. Anal. Biochem., 311: 76-79.
Kawahara S (1998). Chemotherapeutic agents under study. Nippon Rinsho. 56 (12): 3096-3099.
Kelly ME, Batty RA, McIntyre DC (1999). Cortical spreading depression reversibly disrupts convulsive motor seizure expression in amygdale kindled rats. Neurosci., 91: 305-313.
Kelly ME, Staines WA, McIntyre DC (2002). Secondary generalization of hippocampal kindled seizures in rats: examination of the periform cortex. Brain Res., 957(1): 152-161.
Kim J, Ohtani H, Tsujimoto M, Sawada Y (2009). Quantitative comparison of the convulsive activity of combination of twelve fluoroquinolones with five nonsteroidal anti-inflammatory agents. Drug Metab. Pharmacokin., 24(2): 167-174.
Kohanski MA, Dwyer DJ, Hayete B, Lawrence CA, Collins JJ (2007). A common mechanism of cellular death induced by bactericidal antibiotics. Cell, 130: 797-810.
Kohanski MA, Dwyer DJ, Collins JJ (2010). How antibiotics kill bacteria: from targets to networks. Nat. Rev. Microbiol., (8): 423-435.
Kourie JI (1998). Interaction of reactive oxygen species with ion transport mechanisms. Am. J. Physiol., 275: 1-24.
Koziel R, Zablocki K, Duszynski J (2006). Calcium signals are affected by ciprofloxacin as a consequence of reduction of mitochondrial DNA content in Jurkat cells. Antimicrob. Agents. Chemother., 50: 1664-1671.
Koziel R, Szczepanowska J, Magalska A, Piwocka K, Duszynski J, Zablocki K (2010). Ciprofloxacin inhibits proliferation and promotes generation of aneuploidy in Jurkat cells. J. Physiol. Pharmacol., 61(2): 233-239.
Lawrence JW, Darkin- Rattray S, Xie F, Neims AH, Rowe TC (1993). 4-Quinolones cause a selective loss of mitochondrial DNA from mouse L1210 leukemia cells. J. Cell Biochem., 51: 165-174.
Lawrence KR, Adra M, Keir C (2006). Hypoglycemia-induced anoxic brain injury possibly associated with levofloxacin. J. Infect., 52(6): 177-180.
Li Q, Peng S, Sheng Z, Wang Y (2010). Ofloxacin induces oxidative damage to joint chondrocytes of juvenile rabbits: excessive production of reactive oxygen species, lipid peroxidation and DNA damage. Eur. J. Pharmacol., 626 (2-3): 146-153
Liu Q, Wang JC (1999). Similarity in the catalysis of DNA breakage and rejoining by type IA and IIA DNA topoisomerases. Proc. Natl. Acad. Sci. USA, 96: 881- 886.
Liu H, Mulholland SG (2005). Appropriate antibiotic treatment of genitourinary infections in hospitalized patients. Am. J. Med., 118 (7A): 14S-20S.
Lowry OH, Rosebrough NJ, Farr AL, Randall RJ (1951). Protein measurement with the Folin phenol reagent. J. Biol. Chem., 193(1): 265-275.
MacDougall C, Guglielmo BJ, Maselli J, Gonzales R (2005). Antimicrobial drug prescribing for pneumonia in ambulatory care. Emer. Infect. Dis., 11 (3): 380-384.
Machado FR, Ferreira AG, Da Cunha AA, Tagliari B, Mussulini BH, Wofchuk S, Wyse AT (2011). Homocysteine alters glutamate uptake and Na+,K+-ATPase activity and oxidative status in rats hippocampus: protection by vitamin C. Metab Brain Dis., 26(1): 61-67.
Maeda N, Tamagawa T, Niki I, Miura H, Ozawa K, Watanabe G, Nonogaki K, Uemura K, Iguchi A (1996). Increase in insulin release from rat pancreatic islets by quinolone antibiotics. Brit .J. pharmacol., 117:372-376.
Minami M, Yoshikawa H (1979).A simplified assay method of superoxide dismutase activity for clinical use. Clin. Chim. Acta, 92: 337-342.
Mingeot-Leclercq MP, Tulkens PM (1999). Aminoglycosides: nephrotoxicity. Antimicrob. Agents Chemother, 43: 1003–1012.
Moein S, Farzami B, Khaghani S, Moein MR, Larijani B (2007). Antioxidant properties and prevention of cell cytotoxicity of Phlomis persica Boiss. DARU. 15(2): 83-88.
Motomura M, Kataoka Y, Takeo G, Shibayama K, Ohishi K, Nakamura T, Niwa M, Tsujihata M, Nagataki S (1991). Hippocampus and frontal cortex are the potential mediatory sites for convulsions induced by new quinolones and non-steroidal anti-inflammatory drugs. Int. J. Clin. Pharmacol. Ther. Toxicol., 29(6): 223-227.
Müller M (2000). Microdialysis in clinical drug delivery studies. Adv. Drug Deliv. Rev., 45 (2-3): 255-269.
Munhoz CD, Kawamoto EM, de Sá Lima L, Lepsch LB, Glezer I, Marcourakis T, Scavone C (2005). Glutamate modulates sodium-potassium-ATPase through cyclic GMP and cyclic GMP-dependent protein kinase in rat striatum. Cell Biochem. Funct., 23(2): 115-123.
Nelson JM, Chiller TM, Powers JH, Angulo FJ (2007). Fluoroquinolone-resistant Campylobacter species and the withdrawal of fluoroquinolones from use in poultry: a public health success story. Clin. Infect. Dis., 44 (7): 977-980.
Nilson P, Ronne- Engstrom E, Flink R, Ungerstedt U, Carlson H, Hillered L (1994). Epileptic seizure activity in the acute phase following cortical impact trauma in rat. Brain. Res., 637: 227–232.
Oliphant CM, Green GM (2002). Quinolones: a comprehensive review. Am. Fam. Physician, 65: 455– 464.
Owens RC, Ambrose PG (2005). Antimicrobial safety: focus on fluoroquinolones. Clin. Infect. Dis., 41 (2): 144-157.
Papadoyannis LN, Samanidou VF, Nitsos CHC (1999). Simultaneous determination of nitrite and nitrate in drinking water and human serum by high performance anion-exchange chromatography and UV detection. J. Liquid Chromatogr. Relat. Technol., 22(13): 2023-2041.
Pervaiz S, Clement MV (2002). A permissive apoptotic environment: function of a decrease in intracellular superoxide anion and cytosolic acidification. Biochem. Biophys. Res. Commun., 290: 1145-1150.
Pomposiello PJ, Demple B (2000). Identification of SoxS-Regulated Genes in *Salmonella enterica* Serovar Typhimurium. Bacteriology, 182(1): 23-29.
Rajasekaran K (2005). Seizure-induced oxidative stress in rat brain regions: blockade by nNOS inhibition. Pharmacol. Biochem. Behav., 80(2): 263-272.
Rotruck JT, Pope AL, Ganther HE, Swanson AB, Hafeman DG, Hoekstra WG (1973). Selenium: Biochemical role as a component of glutathione peroxidase. Science, 179: 588-590.
Saito T, Nakamura M, Watari M, Isse K (2008). Tardive seizure and antibiotics: case reports and review of the literature. J. ECT, 24(4): 275-276.
Sawas AH, Gilbert JC (1984). Possible mechanism of inhibition by lipid peroxidation of ATPase activities of rat cerebral cortex synaptosomes. Arch. Int. Pharmacodyn. Ther., 269(1): 4-11
Schmuck G, Schürmann A, Schlüter G (1998). Determination of the excitatory potencies of fluoroquinolones in the central nervous system by an *in vitro* model. Antimicrob. Agents Chemother., 42: 1831-1836.
Segev S, Yaniv I, Haverstock D, Reinhart H (1999). Safety of long-term therapy with ciprofloxacin: data analysis of controlled clinical trials and review. Clin. Infect. Dis., 28: 299-308.
Sen S, Jaiswal AK, Yanpallewar S, Acharya SB (2007). Aniogenic

potential of ciprofloxacin and norfloxacin in rats. Sing. Med. J., 48(11): 1028-1032.
Shelat PB, Chalimoniuk M, Wang JH, Strosznajder JB, Lee JC, Sun AY, Simonyi A, Sun GY (2008). Amyloid beta peptide and NMDA induce ROS from NADPH oxidase and AA release from cytosolic phospholipase A2 in cortical neurons. J. Neurochem., 106: 45-55.
Shimizu T, Numata T, Okada Y (2004). A role of reactive oxygen species in apoptotic activation of volume-sensitive Cl^- channel. Proc. Natl. Acad. Sci. USA. 101: 6770-6773.
Sies H (1991). Oxidative stress: from basic research to clinical application. Am. J. Med., 30; 91 (3C): 31S-38S.
Sies H (1999). Glutathione and its role in cellular functions. Free Radic. Biol. Med., 27(9-10): 916-921.
Sprandel KA, Rodvold KA (2003). Safety and tolerability of fluoroquinolones. Clin. Corn., (3): 29-36.
Sun AY, Chen YM (1998). Oxidative stress and neurodegenerative disorders. J. Biomed. Sci., 5: 401-414.
Tomita T, Onishi M, Sato E, Kimura Y, Kihira K (2007). Gatifloxacin induces augmented insulin release and intracellular insulin depletion of pancreatic islet cells. Biol. Pharm. Bull., 30 (4): 644-647.
Tsakiris S (2001). Effects of L-phenylalanine on acetylcholinesterase and Na^+,K^+-ATPase activities in adult and aged rat brain. Mech. Ageing Dev., 122:491-501.
Umezawa N, Arakane K, Ryu A, Mashiko S, Hirobe M, Nagano T (1997). Participation of reactive oxygen species in phototoxicity induced by quinolone antibacterial agents. Arch. Biochem. Biophys., 342: 275-281.
Ungerstedt U (1991). Microdialysis - Principles and applications for studies in animals and man. J. Intern. Med., 230(4): 365-373.
Vallurupalli S, Huesmann G, Gregory J, Jakoby IV (2008). Levofloxacin-associated hypoglycaemia complicated by pontine myelinolysis and quadriplegia. Dia. Med., 25: 856–859.
Voorn P, Vanderschuren LJ, Groenewegen HJ, Robbins TW, Pennartz CM (2004). Putting a spin on the dorsal-ventral divide of the striatum. Trends. Neurosci., 27: 468-474.
Wardman P, Candeias LP (1996). Fenton chemistry: an introduction. Radiat. Res., 145: 523-531.
Wieloch T (1985). Hypoglycemia-induced neuronal damage prevented by an N-methyl-D-aspartate antagonist. Science, 230: 681–683.
Winterbourn CC (1995). Toxicity of iron and hydrogen peroxide: the Fenton reaction. Toxicol. Lett., 82-83: 969-974.
Yin W, Cheng W, Shen W, Shu L, Zhao J, Zhang J (2007). Impairment of Na^+,K^+- ATPase in CD95(APO-1)-induced human T-cell leukemia cell apoptosis mediated by glutathione depletion and generation of hydrogen peroxide. Leukemia, 21: 1669-1678.