Research Article

Evaluation of the effect of co-administration of resveratrol and vitamin E on carbamazepine-induced oxidative stress in male adult wistar rats

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ABSTRACT

Background: Carbamazepine (CBZ) as a drug used in the treatment of epilepsy and neuropathic pain has been shown to stimulate the effects of free radicals. Resveratrol, known as 3,5,4′-trihydroxystilbene, is found in grapes and other plant product. It effectively scavenges free radicals and other oxidants. Vitamin E is a lipid soluble antioxidant present in all cellular membranes. The present study assessed the combined effect of vitamin E and resveratrol on biomakers of carbamazepine- induced oxidative stress. **Methods:** Adult male Wistar rats (n = 25) were randomly allotted to five groups: Group I (control) received distilled water; Group II received CBZ (50 mg/kg); Group III received CBZ (50 mg/kg) and vitamin E (200 mg/kg); Group IV received CBZ (50 mg/kg) and resveratrol (20mg/kg); Group V received CBZ (50 mg/kg) and the co-administration of vitamin E at 200 mg/kg and resveratrol at 20 mg/kg. Administration was done orally daily for 45 days, after which the animals were sacrificed and sera samples were used for biochemical analyses. The results show that treatment with CBZ significantly (p<0.01) increased malondyaldehyde (MDA) serum level and decreased the levels of oxidative stress bio-makers [superoxide dismutase (SOD), catalase(CAT) and glutathione peroxidase (GPx)] when compared to that of normal control. However, treatment with vitamin E (200mg/kg) and resveratrol (20mg/kg) significantly (p<0.01) reduced CBZ-induced increase in serum MDA level and increased the level of oxidative stress bio-makers (SOD, CAT and GPx) in comparison to CBZ-treated group. The co-administration of vitamin E (200mg/kg) and resveratrol (20mg/kg) showed non-statistically significant increase in SOD, CAT and GPx and reduced serum MDA level in comparison to either vitamin E or resveratrol treated group. The results support that vitamin E 200mg/kg and resveratrol 20mg/kg or their combination ameliorates CBZ-induced oxidative stress in male Wistar rats. The effects of these antioxidants are considered to be related to their intrinsic ability to scavenge free radicals

INTRODUCTION

Carbamazepine (CBZ) is a drug widely used in the control of seizures disorders, relief of neuralgia, and variety of other mental disorders. The mechanism of action of CBZ and its congeners is relatively well understood as it decreases neuronal activity (Malarvizhi

et al., 2012). It stabilizes voltage-gated sodium channels in their inactive state, thereby reducing their open state probability. This leaves the affected cells less excitable until the drug dissociates. CBZ also stimulate gamma amino butyric acid (GABA) receptor agonist that potentiates α_1, β_2, and γ_2 subunits. This mechanism may contribute to its efficacy in neuropathic pain and bipolar disorder (Getinet, 2016). Laboratory study has further shown that it is a serotonin releasing agent and possibly a serotonin reuptake inhibitor (*Kawata et al.*, 2001). However, recent studies have reported that CBZ induces oxidative stress alongside its therapeutic effects (Malarvizhi *et al.,* 2012; Eghbal, *et al.,* 2013). Reactive metabolite of

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CBZ known as arene oxide has been implicated as a mechanism for cellular damages caused by this drug (Bavdekar *et al.*, 2004).

The term "oxidative stress" has been used to define a state in which reactive oxygen species (ROS) and reactive nitrogen species (RNS) reach excessive levels, either by excess production or insufficient removal (Johansen *et al.*, 2005). Thus, it is an "imbalance between oxidants and antioxidants in favour of the oxidants, potentially leading to cell damage". Being highly reactive molecules, the pathological consequence of excess ROS and RNS is damage to proteins, lipids and DNA. Consistent with the primary role of free radicals formation, this oxidative stress damage may lead to physiological dysfunction, cell death, pathologies such as diabetes, cardiovascular diseases and cancer, aging and other neurodegenerative diseases (Ahmad *et al.*, 2015; Geon et al., 2015; Ngoc, 2015).

Whenever a cell's internal environment is perturbed by infections, diseases, toxins or nutritional imbalance, mitochondria diverts electron flow away from itself, forming ROS and RNS, thus lowering oxygen consumption. This "oxidative shielding" acts as a defense mechanism for either decreasing cellular uptake of toxic pathogens or chemicals from the environment, or to kill the cell by apoptosis and thus avoid the spreading to neighboring cells. Therefore, ROS formation is a physiological response to stress (Basir *et al.*, 2005).

Resveratrol (RSV) is a natural polyphenol found in grapes, red wine, peanuts and berries (Chen *et al,* 2016). It is a plant secondary metabolite derived from shikimate-phenylpropanoid and/or polyketide pathway. The plant shikimate pathway has two end-products that are the entry to the biosynthesis of phenylpropanoids: phenylalanine and tyrosine. Resveratrol is a product of the phenylalanine/polymalonate pathway, being the last step of this biosynthetic pathway, it can be synthesized either from phenylalanine or tyrosine precursors which produce *para*-coumaric acid (also known as *para*-hydroxycinnamic acid) (Surajit and Ka-Yun, 2011).

Vitamin E is a member of eight fat-soluble compounds that include both tocopherols and tocotrienols (Maheswari *et al.*, 2015). It is one of the most fascinating natural resources that have potential influence on wide range of mechanisms predisposing humans to health and disease challenges (Catalgol and Ozer, 2011). It is the principal lipid soluble chain-breaking antioxidant in mitochondria, microsomes, and lipoproteins (Maheswari *et al.*, 2015). Vitamin E has been used as an exogenous source of essential, fat soluble nutrient that protects human body from the deleterious effects of free radicals since the body cannot manufacture its own vitamin E (Catalgol and Ozer 2011).

Reports on the antioxidant effect of co-administration of resveratrol and vitamin E on CBZ induced oxidative stress are scanty. Hence the present study aimed at investigating the ameliorative effect of administration of resveratrol and vitamin E singly or combination on carbamazepine induced oxidative stress in adult male Wistar rats.

MATERIALS AND METHODS

Experimental animals

Experimental ethics were in accordance with the guidelines for animal research, as stated in the NIH guidelines for the care and use of laboratory animals (National Academy of Sciences and National Institute of Health Publications, 2011). Twenty five adult male Wistar rats weighing 150-200g were obtained from the Animal House, Department of Human Physiology, Faculty of Medicine, Ahmadu Bello University, Zaria, Nigeria. All the rats were kept in well ventilated steel wire cages with normal photoperiod and fed the same type of feed (Vital Feeds Nigeria plc) with access to water *ad libitum*. The rats were allowed to acclimatize under laboratory conditions for two weeks before commencement of the experiment. The animals were allotted to five groups (n=5). All administrations were done orally daily by gavage for forty five days:

Group I: Distilled water 1ml/kg/day.
Group II: Carbamazepine 50mg/kg/day (Thakur, *et al*., 2012).
Group III: Carbamazepine 50mg/kg/day and vitamin E 200mg/kg/day (Maheswari, *et al*., 2015).
Group V: Carbamazepine 50mg/kg/day and resveratrol 20mg/kg/day (Rai *et al.,* 2013).
Group V: Carbamazepine 50mg/kg/day, and co-administration of vitamin E (200mg/kg/day) and resveratrol (20mg/kg/day).

Chemicals and drugs

These include vitamin E (Greenbrier International, INC. USA), resveratrol (Mega Resveratrol Candlewood Stars Limited. USA), carboxymethylcellulose, carbamazepine (Micro Labs Limited. India), corn oil, and chloroform. The drugs were freshly prepared.

Evaluation of Antioxidant Enzymes and Lipid Peroxidation

The animals were sacrificed under chloroform anaesthesia 24h after last administration. Blood samples (about 3 mL each) were collected via cardiac puncture into plain tubes.The samples were allowed to clot and the serum was separated by

centrifugation using a Denley BS400 centrifuge (England) at 3000 rpm for 10 min.

Malondialdehyde (MDA)
Serum MDA concentration was assayed by the method of Albro *et al.,* (1986) and Das *et al.,* (1990). In brief, 0.1 ml of serum was treated with 2 ml of (1:1:1 ratio) tert butyl alcohol (TBA)– trichloroacetic acid (TCA)– hydrochloric acid (HCl) reagent (TBA 0.37%, 0.25N HCl and 15% TCA) and placed in water bath for 15 min, cooled and centrifuged and then clear supernatant was measured at 535nm against reference blank.

Catalase (CAT)
Catalase (CAT) serum level was measured using Abebi's method (1974). Briefly, 10µl of serum was added to a test tube containing 2.80ml of 50mM potassium phosphate buffer (pH 7.0). The reaction was initiated by adding 0.1ml of freshly prepared 30mM H_2O_2 and the decomposition rate of H_2O_2 was measured at 240nm for 5 min on a spectrophotometer.

Superoxide dismutase (SOD)
Superoxide dismutase (SOD) was determined by the method described by Fridovich (1975). Serum sample of 0.1ml was diluted in 0.9ml of distilled water to make 1:10 dilution of microsome. An aliquant mixture of 0.2ml of the diluted microsome was added to 2.5ml of 0.05M carbonate buffer. The reaction was started with the addition of 0.3ml of 0.3mM Adrenaline. The reference mixture contained 2.5ml of 0.05M carbonate buffer, 0.3ml of 0.3mM Adrenaline and 0.2ml of distilled water. The Absorbance was measured over 30 seconds up to 150 seconds at 480nm.

Glutathione peroxidase
Glutathione peroxidase was determined by the method of Ellman, (1959). Briefly, serum sample of 0.5 ml was precipitated by 2 ml of 5% trichloroacetic acid. Ellman's reagent of 0.5 ml (0.0198% DTNB in 1% sodium citrate) and 3 ml of phosphate buffer (pH 8.0) was added. The colour developed was read at 412 nm.

Statistical Analysis
Data obtained from the study were expressed as mean ± SEM. Statistical analysis was carried out using version 20 of statistical package for the social sciences (IBM Corp. Armonk, NY) with the aid of one way analysis of variance (ANOVA) and Tukey's post-hoc test. Values with ($p<0.05$) were considered significant.

RESULTS
Effect of Vitamin E and Resveratrol on Lipid Peroxidation
The effect of vitamin E and resveratrol on malondialdehyde (MDA) concentration in CBZ-

induced oxidative stress is shown in Figure 1. The result shows that treatment with carbamazepine significantly ($p<0.01$) increased MDA serum concentration to 1.42 ± 0.04µmol/L when compared to that of normal control 0.92 ± 0.04µmol/L. However, treatment with vitamin E and resveratrol significantly ($p<0.01$) reduce CBZ-induced increase in MDA serum concentration to 1.02 ± 0.05µmol/L and 1.00 ± 0.03 µmol/L respectively, in comparison to CBZ-treated group. The result also shows that co-administration of vitamin E and resveratrol reduced CBZ-induced increase in MDA serum concentration (0.98 ± 0.05µmol/L) when compared to either vitamin E or resveratrol treated groups although not statistically significant.
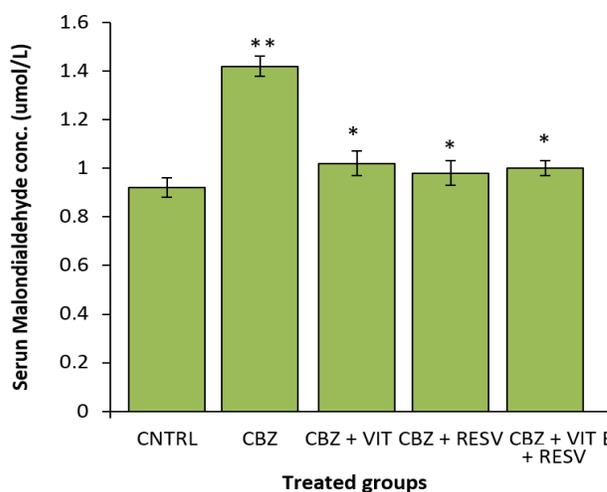

Fig. 1: Serum malondialdehyde concentrations in adult male Wistar rats. CNTRL= control (1 ml/kg of distilled water), CBZ= carbamazepine, Resv= resveratrol, Vit E = vitamin E. Superscripts * and ** indicate statistical significant difference ($p<0.01$) when compared to CBZ and CNTRL treated groups respectively.

Effect of Vitamin E and Resveratrol on Superoxide Dismutase serum level
Figure 2 shows the effect of vitamin E and resveratrol on superoxide dismutase (SOD) serum level in CBZ-induced oxidative stress. The result shows that treatment with carbamazepine significantly (p<0.01) decreased SOD serum level to 2.04 ± 0.02 IU/L in comparison to that of the normal control (2.40 ± 0.03IU/L); however, treatment with vitamin E and resveratrol significantly (p<0.01) increased SOD serum level to 2.42 ± 0.05IU/L and 2.48 ± 0.05IU/L respectively, in comparison to CBZ-treated group. Furthermore, co-administration of vitamin E and resveratrol shows a little increase in SOD serum level to 2.52 ± 0.04IU/L in comparison to either
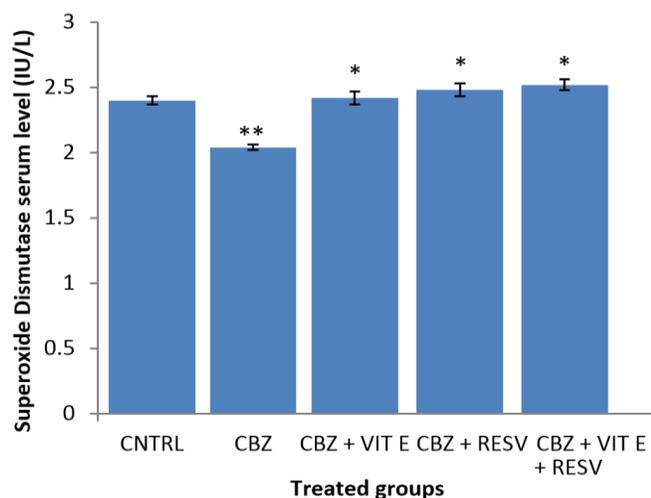
Fig. 2: Serum superoxide dismutase level in adult male Wistar rats. CNTRL= control (1 ml/kg of distilled water), CBZ= carbamazepine, Resv= resveratrol, Vit E = vitamin E. Superscripts * and ** indicate statistical significant difference (*p*<0.01) when compared to CBZ and CNTRL treated groups respectively.

vitamin E or resveratrol treated groups although not statistically significant.

Effect of Vitamin E and Resveratrol on Catalase serum level

The effect of vitamin E and resveratrol on catalase (CAT) serum level in CBZ-induced oxidative stress is shown in Figure 3. The result reveals that treatment with carbamazepine significantly (p<0.01) decreased CAT serum level (46.60 ± 0.40IU/L) in comparison to that of the normal control (52.00 ± 0.32IU/L). However, treatment with vitamin E and resveratrol significantly (p<0.01) increased CAT serum level to (51.80 ± 0.49IU/L) and (53.20 ± 0.37 IU/L) respectively, in comparison to CBZ-treated group. Furthermore, co-administration of vitamin E and resveratrol reveals an increase in CAT serum level 54.00 ± 0.31IU/L when compared to either vitamin E or resveratrol treated group

Effect of Vitamin E and Resveratrol on Glutathione Peroxidase serum level

Figure 4 shows the effect of vitamin E and resveratrol on glutathione peroxidase (GPx) serum level in CBZ-induced oxidative stress. The result shows that treatment with carbamazepine significantly (P<0.01) decreased GPx serum level to 42.20 ± 0.49IU/L in comparison to that of the normal control 45.20 ± 0.37IU/L. Treatment with vitamin E and resveratrol significantly (p<0.01) increased GPx serum level to 47.60 ± 0.40 IU/L

and 48.80 ± 0.37 IU/L respectively, in comparison to CBZ-treated group. Furthermore, co-administration of vitamin E and resveratrol increased GPx serum level to 49.20 ± 0.66 IU/L in comparison to either vitamin E or resveratrol treated group.
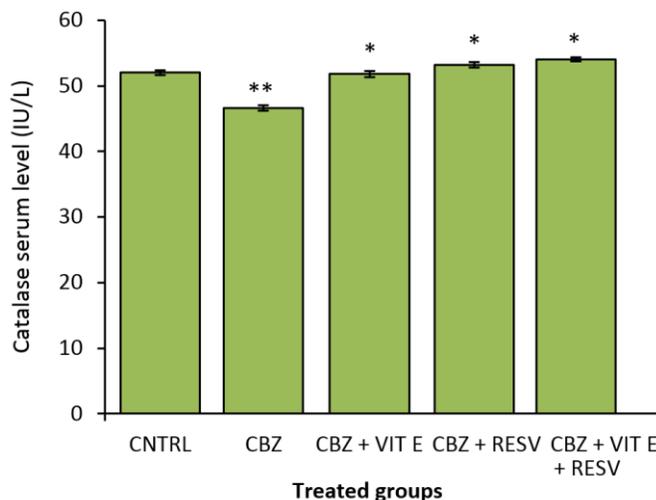

Fig. 3: Serum catalase level in adult male Wistar rats. CNTRL= control (1 ml/kg of distilled water), CBZ= carbamazepine, Resv= resveratrol, Vit E = vitamin E. Superscripts * and ** indicate statistical significant difference (*p*<0.01) when compared to CBZ and CNTRL treated groups respectively.
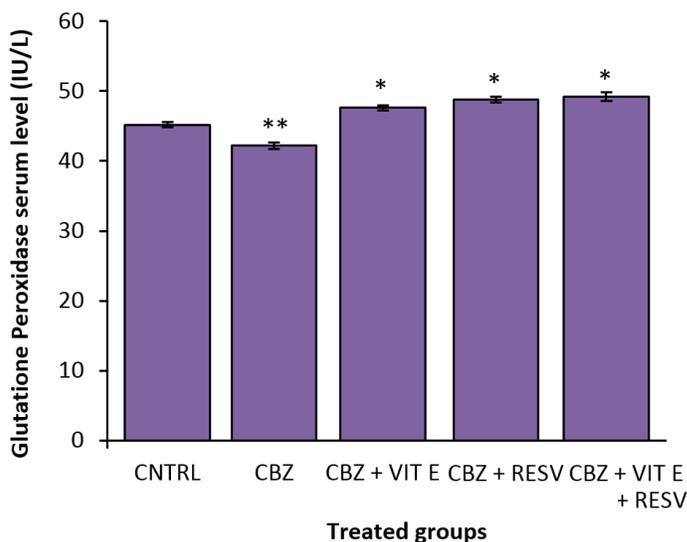

Fig. 4: Serum Glutathione peroxidase level in adult male Wistar rats. CNTRL= control (1 ml/kg of distilled water), CBZ= carbamazepine, Resv= resveratrol, Vit E = vitamin E. Superscripts * and ** indicate statistical significant difference (*p*<0.01) when compared to CBZ and CNTRL treated groups respectively.

DISCUSSION

In recent years, there has been increased interest in the toxicological aspect of free radicals with

studies paying attention on various mechanistic damages of oxidative stress and the physiological responses from various cells, tissues and organ components. Carbamazepine known as a broad spectrum agent for the treatment of psychiatric and neurological disorders, is reported to stimulate the activities of free radicals through secondary metabolites such as arene oxide thus resulting in cellular damages (Eghbal *et al.,* 2013). MDA is an end product and remnant of lipid peroxidation which acts as a cytotoxic messenger for primary reactions. It can escape from cells to initiate attack in other parts of the body (Ememe *et al.,* 2015). In the present study, treatment of Wistar rats with carbamazepine significantly (p<0.01) increased serum MDA level when compared to normal control, however, treatment with resveratrol and vitamin E singly or combination reduced CBZ-induced increase in serum MDA level (Figure 1). Our findings are consistent with the report of Imad, (2012), Thakur *et al.,* (2012) and Maheswari *et al.,* (2015); all of whom reported elevated serum MDA levels after carbamazepine treatments in experimental models. The rise in the level of plasma lipid peroxidation may be due to secondary metabolites formed during carbamazepine administration. Free radicals in the absence of an efficient defense mechanism can initiate peroxidation of membrane poly unsaturated fatty acids thus fragmenting it into alkanes and aldehydes (Thakur *et al.,* 2012). Lipid peroxidation is said to occur in areas where polyunsaturated fatty acid side chains are prevalent (Thakur *et al.,* 2012). These chains react with O_2 creating the peroxyl radical, which can obtain H^+ from another fatty acid, creating a continuous reaction. Pushpalatha *et al.,* (2013) and Xiao (2015) both reported significant decrease in MDA level after scopolamine induced cognitive deficits and strenuous exercise in rats were treated with resveratrol respectively. The reduction in MDA level was previously attributed to scavenging activity of resveratrol on membrane lipid peroxyl radical (Ememe *et al.,* 2015). Resveratrol as a polyphenolic compound is rapidly metabolized in the liver and intestine of rats through glucuronidation and sulphonation, thus the bioavailability is high, as approximately 90% is reported to be present in the plasma (Ramprasath and Jones, 2010). Vitamin E on the other hand is known for its radical chain braking antioxidant

properties (Bjelakovic *et al.,* 2007; Maya *et al.,* 2012). The decrease in MDA after vitamin E supplementation could be due to prevention of chain initiation and propagation of free radical reaction and lipid peroxidation in cellular membrane. It acts through glutathione peroxidase pathway where it reacts with free radicals produced in the lipid peroxidation resulting in protection of the cell membrane against oxidation (Traber and Atkinson, 2007). Our findings also corroborate the works of Salama *et al.,* (2013) and Maheswari *et al.,* (2015).

There are several mechanisms used to counteract oxidative stress; by producing endogenous or exogenous antioxidants. The roles of antioxidants are to neutralize excess free radicals and protect tissue damage (Chuong *et al.,* 2008). Endogenous antioxidant molecules include various enzymes such as SOD, GPx and CAT as well as other non-enzymatic molecules (Geon *et al.,* 2015). The first line of defence against free radicals is SOD, which catalyzes the dismutation of superoxide anion radical into hydrogen peroxide (H_2O_2). Subsequently, H_2O_2 is transformed into water and oxygen by CAT or GPx. Glutathione peroxidase enzyme removes H_2O_2 by using it to oxidize reduced glutathione into oxidized glutathione (Chuong *et al.,* 2008). In certain disease conditions, the antioxidant systems can be overwhelmed (Birben *et al.,* 2012). In the present study treatment with carbamazepine significantly (p<0.01) decreased SOD, CAT and GPx serum level in comparison to that of the normal control, treatment with resveratrol and vitamin E singly or combination increased serum SOD, CAT and GPx levels as shown in Figures 2, 3 and 4 respectively. The decreased serum levels of these enzymes by carbamazepine could be the result of excessive utilization or the consistent depletion of these endogenous antioxidant enzymes pool in trying to ameliorate or combat the existing free radicals and their adverse effects. The results of our findings also agree with several clinical and experimental works which show that CBZ can deplete endogenous antioxidant system (Thakur *et al.,* 2012; Egbal *et al.,* 2013). The increased serum levels of antioxidant enzymes by resveratrol is in conformity with the works of Pushpalatha *et al.,* (2013) and Ememe *et al.,* (2015); both of whom have shown that resveratrol can upregulate depleted antioxidant enzymes. The antioxidant

effect of resveratrol is attributed to the blockade of radical propagation by formation of a resonance-stabilized peroxy radical (Kovacic and Somanathan, 2010). Resveratrol can also scavenge hydroxyl radicals with a reaction rate constant of 9.45×10^8 m^{-1}sec^{-1} (Das and Maulik, 2006). Moreover, resveratrol was shown previously to inhibit the activities of enzymes involved in production of ROS such as nicotinamide adenine dinucleotide phosphateoxidase (Chow *et al.,* 2007), hypoxanthine/xanthine oxidase, (Shigematsu *et al.,* 2003) and myeloperoxidase (Kohnen *et al.,* 2007). In addition to these, the lipophilic character of resveratrol may have enabled it bind to lipoprotein particles thereby improving its antioxidant activity (Belguendouz *et al.,* 1998). Vitamin E is an important free radical scavenger which exerts non enzymatic protection against lipid peroxidation; it plays a protective role during oxidative stress by preventing oxidative processes from progressing (Maheswari *et al.,* 2015) and can also attenuate glutathione levels (Al Hashem, 2009).

CONCLUSION

The present result has shown that administration of resveratrol 20mg/kg and vitamin E 200mg/kg singly ameliorated carbamazepine induced oxidative stress in male Wistar rats. However co-administration of resveratrol and vitamin E did not show statistical significant change when compared to either resveratrol or vitamin E treated groups.

REFERENCE

Abebi, H. (1974). Catalase. Methods in enzymatic analysis. Vol 2, New York Acadamic press, 674-684.

Ahmad, N.K., Mushtaq, A., Nadia, M. and Rahmat, A.K. (2015). Role of Antioxidant in Oxidative Stress and Diabetes Mellitus. *Journal of Pharmacognosy and Phytochemistry*. 3:(6), 217-220

Albro, P.W., Corbelt, J.T. and Schroeder, J.L. (1986).Application of the thiobarbiturate assay to the measurement of lipid peroxidation products in microsomes. *Chemistry and Biology Interaction, 86*, 185 – 194

AlHashem, F. (2009). Camel milk protects against aluminium chloride-induced toxicity in the liver and kidney of white albino rats.

American Journal of Biochemistry and Biotechnology, 5: 98-108.

Basir, S.F., Baquer, N.Z., Hussain, M.E., Moorthy, K., Siddiqui, M.R. and Taha, A. (2005).Amelioration of Altered Antioxidant Status and Membrane-linked Functions by Vanadium and *Trigonella*in Alloxan Diabetic Rat Brains. *Journal of Bioscience*, 30:483–490

Bavdekar, S. B., Muranjan, M. N., Gogtay, N. J., Kantharia, V. and Kshirsagar, N. A. (2004). Anticonvulsant hypersensitivity syndrome:lymphocyte toxicity assay for the confirmation of diagnosisand risk assessment. *Annals of Pharmacotherapy,* 38(10):1648-50.

Belguendouz, L., Fremont, L. and Gozzelino, M.T. (1998). Interaction of transresveratrol with plasma lipoproteins. *Biochemistry Pharmacology*, *55*: 811–816

Birben, E., Sahiner, U.M., Sackesen, C., Erzurum, S. and Kalayci, O. (2012). Oxidative Stress and Antioxidant Defense. *World Allergy Organization Journal,* 5: 9-19.

Bjelakovic, G., Nikolova, L.L., Gluud, S. and Gluud, C. (2007). Mortality in randomized trials of antioxidant supplements for primary and secondary prevention. *Journal of the American Medical Association,* 297(8): 842-857.

Catalgol, B. and Ozer N.K (2011). Protective effects of vitamin E against hypercholesterolemia- induced age-related disease. *Genes and Nutrition,* 1-8. doi 10.1007/s12263-011-0235-9.

Chen, W.M.,Shaw, L.H.,Chang, P.J., Tung, S.Y.,Chang, T.S.,Shen,C.S.,Hsieh,Y.Y. and Wei, K.L (2016). Hepatoprotective effect of resveratrol against ethanol-induced oxidative stress through induction of superoxide dismutase *in vivo* and *in vitro. Experimental and Therapeutic Medicine*, 11: 1231-1238.

Chuong P.H., Lien, A.P., and Hua, H. (2008). Free Radicals, Antioxidants in Disease and Health. *International Journal of Biomedical Science,* 4(2): 89-96.

Chow, S.E., Hshu, Y.C., Wang, J.S. and Chen J.K. (2007). Resveratrol attenuates oxLDL-stimulated NADPH oxidase activity and protects endothelial cells from oxidative functional damages. *Journal of Applied Physiology,*102(4):1520–152760

Das, D.K. and Maulik, N. (2006). Resveratrol in cardioprotection: a therapeutic promise of

alternative medicine. *Molecular Interventions,* 6(1): 36-47.

Das, B.S., Turnham, D.I., Painack, J.K., Das, D.E., Satpathy, R. and Base, T.K. (1990). Increased plasma lipid peroxidation in riboflavin deficient malaria-inflected children. *Annual Journal of Clinical Nutrition, 51*, 859- 863.

Eghbal, M.A., Taziki, S., Sattari, M.R. (2013). Protective role of melatonin and taurine against carbamazepine-induced toxicity in freshly isolated rat hepatocytes. *International Journal of Morphology,*31:1081–1089.

Ellman, G.L. (1959). Tissue sulphydryl groups. *Archives of Biochemistry and Biophysics, 82*, 70-77.

Ememe, M.U., Mshelia, W.P. and Ayo, J.O. (2015). Ameliorative effects of resveratrol on oxidative stress biomarkers in horses. *Journal of Equine Veterinary Science,* xx:1-6. http://dx.doi.org/10.1016/j.jevs.2015.02.06.

Fridovich, l. (1975). Superoxide Dismutases. *Annual Review of Biochemistry, 44*, 147-159

Geon, H. K., Jieun, E., Kim, S. J. R. and Sujung, Y. (2015).The Role of Oxidative Stress in Neurodegenerative Diseases. *Experimental Neurobiology* 24(4); 1-16

Getinet Ayano. (2016). Bipolar disorders and carbamazepine: pharmacokinetics, pharmacodynamics therapeutic effects and indications of carbamazepine: review of articles. *Journal of neuropsychopharmacology and mental health*, DOI:10.4172/2472-095X.1000112.

Imad, A. Thanoon., Othman A. Pachachi., and Mohammed M. Al-Sheikh. (2012). Effects of carbamazepine on serum leptin, insulin levels and oxidative stress in epileptic patients.*Annals of the College of Medicine Mosul*, 38 (1): 40-45

Johansen, J.S., Harris, A.K., Rychly, D.J., and Ergul, A. (2005). Oxidative stress and the use of antioxidants in diabetes: linking basic science to clinical practice. *Cardiovascular diabetol, 4(1)5.*

Kawata, Y., Okada, M., Murakami, T., Kamata, A., Zhu, G. and Kaneko, S. (2001). Pharmacological discrimination between effects of carbamazepine on hippocampal basal evoked serotonin release. *British Journal of Pharmacology, 133 (4): 557–567.*

Kohnen, S., Franck, T., Van Antwerpen, P., *et al.,* (2007). Resveratrol inhibits the activity of equine neutrophil myeloperoxidase by a direct interaction with the enzyme. *Journal of Agriculture and Food Chemistry*, 55(20):8080–8087.

Kovacic, P., Somanathan, R. (2010). Multifaceted approach to resveratrol bioactivityfocus on antioxidant action, cell signaling and safety. *Oxidative Medicine and Cellular Longevity*, 3: 86–100.

Maheswari, E., Saraswathy, R.G. and Santhranii, T. (2015). Influence of vitamin E on hepatotoxicity and oxidative stress. *International Journal of Research in Pharmacy and Biosciences,* 2(3): 30-38.

Malarvizhi, A., Kavitha, C., Saravanan, M. and Ramesh, M. (2012). Carbamazepine (CBZ) induced enzymatic stress in gill,liver and muscle of a common carp, *Cyprinus carpio. Journal of King Saudi University–Science*, 24:179–186.

Maya, W., Mayur, K. and Ashar, S. (2012). Pharmaceutical profile of alpha-tocopherol: A brief review. *International Journal of Chemistry and Pharmaceuticals Science,* 1(3): 674-682.

Ngoc, A. L. (2015) Lipoprotein-Associated Oxidative Stress: A New Twist to the Postprandial Hypothesis, *International Journal of Molecular Science,* 16:401-419

Pushpalatha, B., Venumadhav, N., Swathi, M. and Raju, A.B. (2013). Neuroprotective effect of resveratrol against scopolamine-induced cognitive impairment and oxidative stress in rat**.** *Archieves of Biological Science*, 65 (4), 1381-1386.

Rai, A. R., Sampath, M. L., Prabhu, V.,Vasudha, V., Sudhanshu, S. and Gayathri, R. (2013). Resveratrol reverses the restraint stress-induced cognitive dysfunction involving brain antioxidant system in rats. *International Journal of Pharmacy and Pharmaceutical Science, 6*(1): 768-772

Ramprasath, V.R. and Jones, P.J.H. (2010). Anti artherogenic effect of resveratrol. *European Journal of Clinical Nutrition,* 64:660-668.

Shigematsu, S., Ishida, S., Hara, M., *et al.* (2003). Resveratrol, a red wine constituent polyphenol, prevents superoxide-dependent inflammatory responses induced by ischemia/reperfusion platelet-activating factor, or oxidants. *Free Radical Biology and Medicine,*34(7):810–817.

Salama, A.F., Kasem, S.M.,Tousson, E. and Elsisy, M.K. (2013). Protective role of L-carnitine and vitamin E on the testis of herosclerotic rats. *Toxicology and Industrial Health,*1-8.

Surajit D. and Ka-Yun, N.g. (2011).Quantification of *trans*-resveratrol in rat plasma by a simple and sensitive high performance liquid chromatography method and its application in pre-clinical study. *Journal of Liquid Chromatography and Related Technologies, 34 (14)*:1399-1414.

Thakur, S., Maheswari, E. and Saraswathy, G. R. (2012). Amelioration of carbamazepine induced oxidative stress and hematotoxicity by vitamin C. *Spatula DD -Journal on Complementary Medicine and Drug, 2*(3): 173-180.

Traber, M.G. and Atkinson, J. (2007). Vitamin E, antioxidant and nothing more. *Free Radical Biology and Medicine,* 43(1): 4-10.

Xiao, N.N. (2015). Effects of resveratrol supplementation on oxidative damage and lipid peroxidation induced by strenuous exercise in rats *Biomolecules and Therapeutics,*23(4):374-378.