Available online at http://www.journalcra.com

International Journal of Current Research
Vol. 8, Issue, 11, pp.41996-42002, November, 2016

INTERNATIONAL JOURNAL OF CURRENT RESEARCH

ISSN: 0975-833X

RESEARCH ARTICLE

PROTECTIVE EFFECT OF CURCUMIN ON LINDANE-INDUCED NEPHROTOXICITY IN MALE WISTAR RATS

[1]Neelam Yadav, [2]Harendra Kumar and [3,*]Satish Chandra

[1]Department of Zoology, Institute of Basic Sciences, Bundelkhand University, Kanpur Road, Jhansi- 284128, India.
[2]Department of Pathology, S.N. Medical College, Agra-282002, U.P., India
[3]Department of Zoology, Charak Institute of Education, IIM Road, Maura, Hardoi Sitapur By Pass, Lucknow-226020, U.P., India

ARTICLE INFO

Article History:
Received 14th August, 2016
Received in revised form
28th September, 2016
Accepted 20th October, 2016
Published online 30th November, 2016

Key words:

Curcumin,
Lindane,
Renal,
Wistar rats.

ABSTRACT

Lindane; an organochlorine pesticide has been used in agriculture and domestic purposes for several years. The aim of present study was to analyze the oxidative effect of lindane which caused biochemical and ultrastructural changes in adult male wistar rats and to evaluate the possible protective effect of curcumin. Tissues damage was assessed by histopathological observation. Curcumin plays an important role as an antioxidant and is consequently expected to protect tissues from damage caused by reactive oxygen metabolites. Rats were divided into seven goups. Group-A, was given normal diet and water ad libitum. Lindane (30 mg/kg body weight) was administered orally for 14 and 28 days in group- B and group-C respectively. Curcumin (100 mg/kg body wt) was given to Group-D and Group-E. Lindane (30 mg/kg body wt) along with curcumin (100 mg/kg body wt) was administered orally for 28 days in group-F. Group-G, was allowed to metabolized after 14 days of exposure to lindane. Lindane administration lead to a significantly ($p<0.001$) increase in renal lipid peroxidation associated with reduction in levels of GSH, activity of SOD, CAT and GST. Pre-feeding and post-feeding of curcumin resulted in decreased renal levels of lipid peroxides and increased GSH, SOD, CAT and GST activities. Results revealed that curcumin in combination with lindane partially or totally alleviated its toxic effects on the studied parameters. In conclusion, Curcumin have beneficial effects and could be able to antagonize lindane toxicity.

Citation: Neelam Yadav, Harendra Kumar and Satish Chandra, 2016. "Protective effect of curcumin on lindane-induced nephrotoxicity in male wistar rats", *International Journal of Current Research*, 8, (11), 41996-42002.

INTRODUCTION

Lindane is a γ-isomer of hexachlorocyclohexane, has widely used as an organochlorine pesticide and spread in the environment due to its long life time (Wauchope *et al*., 1992). Pesticide extensively employed for public health as well as agricultural purposes in developing countries. Due to its widespread use, lindane widely distributed in ecosystem and become in the form of global pollutant. Several studies have revealed the presence of lindane above permissible limit in body fat, blood, milk and food commodities both in India and abroad (Banerjee *et al.,* 1997; Samanta *et al.,* 1999). It's widely used therapeutic as scabicide, pediculicide and ecto-parasiticide (Fidan *et al*., 2008). Presently lindane also used in lotions, creams and shampoos for the control of lice and mites in humans (Safe, 1993; Budavari *et al*., 1989). Compounds of this chemical class have very low water solubility but are highly soluble in lipids and bio-accumulate (Murphy, 1986). Toxic effects of lindane in mammals include convulsions, ataxia, prostration, damage to fatty tissues and inhibition of sperm motility in sea urchins (Nelson, 1990; Murphy, 1986). It has been reported to induce oxidative stress by interacting with the cell membrane, triggering the generation of reactive oxygen species (ROS) and altering the level of antioxidant molecules which in turn cause severe physiological dysfunction in various organ systems (Barros *et al*., 1991; Bano and Bhatt, 2007). Recent studies indicate that pesticide intoxication produce oxidative stress by the generation of free radicals (Banerjee *et al*., 1999) and induce tissue lipid peroxidation (Yavuz, *et al.,* 2005). ROS arise as by-products of normal cellular metabolism or may be the consequence of exposure to certain chemicals (Kerr *et al*., 1996; Moslen, 1994) and responsible for structural and functional alterations in cells (Fernandez *et al*., 2003; Comporti, 1989; Kappus, 1987). Normal cellular functions depend on a balance between ROS produced and antioxidant defense mechanisms present in the cell. In search for these new

Corresponding author: Satish Chandra
Department of Zoology, Charak Institute of Education, IIM Road, Maura, Hardoi Sitapur By Pass, Lucknow-226020, U.P., India.

chemical entities as modulators of xenobiotic metabolism, we searched literature on Ayurvedic medicinal plants. Several medicinal plants or their active principles have been used as antioxidants and in reducing the toxicity of xenobiotics. However, it based on the experience of traditional system of medicine from different ethnic societies. The medicinal plant *Curcuma longa* (Turmeric) has attracted the interest of research community due to its number of pharmacological activities (Ammon and Whal, 1991; Srimal, 1997). Curcumin, an active component of turmeric (Curcuma longa linn) exhibits an antioxidant property. It is a yellow colored phenolic pigment yielded from the rhizome of turmeric (family Zingiberaceae). Earlier studies have shown that it is an effective antioxidant against oxidative tissue damage and inhibits ROS production (Quiles *et al*., 2002) both, in vitro and in vivo (Joe and Lokesh, 1994), also acts as a scavenger of free radicals (Khanna, 1999). Therefore, the present study has been undertaken to evaluate the ameliorating effect of curcumin on lindane induced biochemical and histopathological alterations in renal tissues of rat.

MATERIALS AND METHODS

Laboratory Animals

Forty-two male wistar rats (weighing 130–150 g) were obtained from the animal house of the IITR (Industrial Institute of Toxicology Research). Animals were caged in seven groups (each group having six rats) and given food & water ad libitum. The animal room was maintained at 21–24 0C and 40–60% relative humidity with 12-h light–dark cycles, the light cycle coinciding with the day light hours. After 2 weeks of acclimation, the groups were assigned at random to one of the following treatments: group A served as control, while groups B and C were treated with 30 mg lindane/kg body weight, up to 14 and 28 days respectively. Group D received curcumin 100 mg/kg body wt up to 14 days than received lindane 30 mg/kg body wt up to next 14 days. Group E was given lindane 30 mg/kg body wt up to 14 days than received curcumin 100 mg/kg b.wt up to next 14 days. While group F was given lindane (30 mg/kg body wt) plus curcumin (100 mg/kg body wt). The animal G received lindane 30 mg/kg body wt up to 14 days and then were kept for metabolism up to next 14 days. The dose of LD50 (lindane), when administered orally to rats has been reported to be given at the rate of 88 mg/kg body wt (Thomas B. Gaines., 1959). Animals were treated orally with the tested compounds every other day for 28 days. The doses of lindane and curcumin were calculated according to the animal's body weight before treatment.

Chemicals

Pure lindane 99.6% & curcumin were purchased from Sigma Aldrich (st. Louis, Mo. USA). All other chemicals were of AR grade and purchased locally.

Sample collection

Wister Rats of each group were killed by decapitation at the end of the treatment period. Samples (kidney) were collected from the sacrificed animals and placed immediately on -20 ºc temperature. Frozen kidney samples were thawed and 200mg of samples was weighed and taken in 2ml of ice –cold saline for enzyme estimation. An amount of 200 mg of sample was weighed separately and taken in 2 ml of 0.02 M EDTA for GSH estimation. Tissue sections were taken from kidneys for histopathological examination, which were fixed in 10% formalin. Organ homogenates were prepared using tissue homogenizer (IKA, Germany) under ice-cold condition. The homogenate was centrifuged for 10 min at 3000 rpm. The supernatant was used for following biochemical estimation.

Measurement of malondialdehyde

Malondialdehyde occurs in lipid peroxidation, measured in kidney tissues after incubation at 95 °C with thiobarbituric acid in aerobic conditions (pH 3.4). The pink colour produced by these reactions was measured spectrophotometrically at 532 nm to measure MDA levels (Shafiq-u-Rehman., 1984). Specific activity was defined as nanomole per milligram protein.

Measurement of Superoxide dismutase (SOD)

SOD was estimated as per the method described by Madesh and Balsubramaniam (1989). It involved generation of superoxide by pyrogallol auto oxidation and inhibition of superoxide- dependent reduction of the tetrazolium dye MTT [3-(4-5dimethyl thiazole 2-xl) 2, 5 dipyenyl tetrazolium bromide] to its formazan, measured at 570nm. The reaction was terminated by the addition of dimethyle sulfoxide (DMSO), which helps to solublize the formazan formed. The colour evolved to stable for many hours and is expressed as SOD units (have been expressed as U/g of protein) [one unit SOD is amount (µg) of protein required to inhibit the MTT reduction by 50%].

Measurement of Catalase (CAT)

Activities of catalase in kidney homogenate were estimated by the method of Begrmeyer (1983), Diluted (1:10) of homogenate was used for estimation of catalase. The optical density was recorded at every 10 sec for 1 min at 240 nm against water blank.

Measurement of Glutahione-s-transferase (GST)

GST activity was measured by the method of Habig *et al.,* 1974. Assay for the activity of GST is based on the spectrophotometric determination of a CDNB (1-Chloro2, 4-Dinitrobenzene) conjugate formed with glutathione in a GST coupled. The conjugate formation is GST catalyzed and therefore is a measure of GST activity. The changes in absorbance were recorded at 340 nm and the enzyme activity was calculated as nmol CDNB conjugate formed /min/mg/protein using a molar extinction coefficient of 9.6x103 M-1 cm-1.

Measurement of Reduced glutathione (GSH)

Reduced glutathione was determined by the method of Jollow *et al.,* (1974). The method described is based on the development of a yellow color when 5, 5'-dithiobis-(2-nitro benzoic acid) Ellan's reagent (DTNB) is added to sulpohydryl compounds. The color develops is fairly stable for 10 minutes. The reaction is read at 412 nm.

Histopathological examination

Small representative pieces (5 mm thickness) of respective organs viz., kidneys were collected in 10% neutral buffered

formalin solution. After 3-4 days fixation, the tissues were trimmed to 2 mm thickness by sharp blade. Further processing was done by dehydrating the tissue in ascending grades of ethyl alcohol, clearing in xylene then tissue is embedded into melted paraffin wax (melting point 58 ^{0}C), after hardening of wax, tissue blocks were prepared & sections were cut with microtome to obtain 4-5 µ thick sections These sections were hydrated by treating them with descending grades of alcohol and water. These tissues were double stained with hematoxyline and eosin stain. Sections were treated with xylene to remove water & mounted with cover slip using DPX as mounting media. Sections were examined with a NIKON (eclips 8i) DXM 1200X light microscope (Japan).

Statistical analysis

All data were analyzed via ANOVA using Graph Pad in Stat Software Inc., v. 3.06, San Digeo, USA followed by Tukey tests and the statistical significance was considered at $P<0.05$.

RESULT AND DISCUSSION

Oxidative damage primarily occurs through the production of reactive oxygen species (ROS) including hydroxyl radicals and hydrogen peroxide that subsequently react with biological molecules, causing damage to membranes and other tissues (Banerjee *et al*., 1999). The present study reports that curcumin ameliorates the lindane induced toxicity. In agreement with previous studies we have shown that lindane induced renal damage in exposed to male rats (Videla, *et al*., 1990) and that this may be due to depletion in cellular thiol (SH) levels (Muller, 1986). Increased generation of superoxide radicals leads to oxidation and depletion of GSH with a lipid peroxidative response. Glutathione, an endogenous antioxidant plays a critical role in detoxification of reactive oxygen species and free radicals. Several studies using animal model have shown that the use of phytochemicals from plant extracts were protective against the oxidative stress induced by many toxic agents mostly by modulating the GSH and GST levels (Shanmugarajan *et al*., 2008; Nandave *et al*., 2007; Amin' 2008; Sarhan *et al*., 2007). GST catalyses the reaction between the thiol (SH) group of GSH and potential alkylating agents, such as lindane, thereby neutralizing the electrophilic sites and rendering them more water soluble. This enzyme is therefore a major component of the GSH redox cycle. The activity of this enzyme is a crucial factor in determining the sensitivity of cells to broaden the range of toxic chemicals. The present study was in agreement with these previous studies as the presence of curcumin elevates GSH and GST levels in the presence or absence of lindane and this may be responsible for the protective effect of the curcumin against lindane toxicity.

Several enzymes in renal tissues have long been considered as effective biochemical markers to understand the early injury. Table-1 depicts the enzymatic antioxidants activities in the renal tissues of control and experimental rats. The present study revealed that the administration of lindane resulted in significant ($P<0.001$) rise in renal lipid peroxidation. Lipid peroxidation is a free redical mediated chain reaction which can be initiated by hydroxyl radicals and attack polyunsaturated fatty acids in membranes resulting in oxidative damage (Hfaiedh *et al.,* 2012).. Which are agreement with previous observation that lindane exposed rat have shown marked increase lipid peroxidation in kidney (Anilakumar *et al.:* 2006). It has been shown that lindane interacts with cell membranes resulting in lipid peroxidation (Fong, *et al*., 1973). Padma *et al*. (2011) also show that lindane elevates lipid peroxidation in rat's renal tissue exposed to lindane toxicity and this was attributed to decrease antioxidant activities. The rise in lipid LPO level may be due to the increase in generation of the free radicals. These free radicals attack cell structure with the body causing damage to cell membrane and enzyme system (Fong, *et al*., 1973). In this regard, Vijayaval *et al.,* (2006) documented that free radical play a prominent role in elevating LPO and potentially leading to cellular damage. This result is in accordance with previous studies using lindane and other pesticides (Koner *et al*., 1998 and Banerjee *et al*., 2001). Induction of cytochrome P450 and other microsomal enzyme by various pesticides, e.g. carbamate, has been reported and it is possible that lindane mediated free radical generation could be through induction of these enzyme (Hayes, 1982, Puatanochokchai *et al*., 2006 and Padma *et al.,* 2012).

Previous studies have shown increase malondieldihyde level in tissues of animals exposed to toxic agents and this effect was attenuated by the use of various plant extracts (Guldur *et al.,* 2010; Al Rejaie, 2009; Nur Azlina *et al*., 2009; Mtgapor and Fazlina, 2006; Iyawe *et al.,* 2006).The dietry intake of these extracts is considered to be relatively safe and without undesirable side effects (Xavier *et al.,* 2004). The increase in renal LPO content produced by lindane was significantly ($P<0.001$) lowered by Pre and post feeding of curcumine. The result obtained from this present study correlates with the previous findings. The study showed that the pre and post – treatment of curcumin attenuates MDA level in the presence of lindane. This reduction in MDA level may be as a result of increase GSH and GST activities in the presence of curcumin. These results are similar to the observation of another study where gallic acid was shown to decrease LPO level in carbon tetrachloride induced damage in albino rats (Jadon *et al*., 2007). Similarly, El-Demerdash *et al*., (2009); Bishnoi *et al*., (2008) have been reported that treatment with curcumin reduced the level of LPO and induced the activities of the antioxidant enzymes, and the levels of SH-groups in sodium arsenite and haloperidol respectively. Renal GSH level was significantly ($P<0.001$) reduced on lindane administration compared to control. The observed decreased GSH level in both renal tissue may be due to utilization of non-protein thiols by increase ROS under lindane induced oxidative stress (Bano and Bhatt, 2007; Pompella *et al.*, 2003; Ahmad *et al.,* 2008). SOD and CAT activities in renal tissues were decreased significantly ($P<0.001$) in lindane exposed groups compared to control **(**Table-1**).** Superoxide dismutase (SOD) and Catalase (CAT) are two important enzymatic antioxidants that act against toxic oxygen free radicals such as superoxide ($O2^-$) and hydroxyl ions (OH^-) in biological systems (Burton *et al*., 1983; Zlko *et al.,* 2002; Weydert *et al.,* 2006). CAT prevents oxidative hazards by catalyzing the formation of H2O and O2 from H2O2 (Kumar and Kuttan, 2003). In this regard, Padma *et al.,* (2011) have documented that lindane cause depletion in activity of CAT in kidney of female Wistar rats. A previous study by Anila Kumar *et al*. (2009) has shown that lindane exposure usually decreases the activities of SOD and CAT. This implies that lindane causes an increased intracellular accumulation of H2O2 and superoxide radicals. The accumulation will further contribute to the membrane damage via lipid peroxidation. In our present study, we observed a decrease in the SOD and CAT activities of the kidney tissues on lindane administration.

Table 1. Kidney

Parameters	Group A	Group B	Group C	Group D	Group E	Group F	Group G
LPO	37.27 ± 0.099	48.20±0.274***	53.30±0.237***	39.10 ± 0.392*	41.75 ± 0.152**	46.93 ±0.286***	47.82 ±0.071***
GST	1.70 ±0.003	0.51 ±0.001***	0.40 ± .001***	1.11± 0.003●	1.01 ±0.002●	0.89 ±0.006 **	0.53 ± 0.00***
GSH	32.11±0.000	24.86± .000***	18.98±0.000***	30.99 ± 0.000*	28.10 ± 0.001**	27.14 ± 0.000**	24.92±0.000 ***
CAT	66.78±0.505	55.93±0.248***	42.98±0.301***	64.95±0.131*	60.97 ±0.221**	58.99±0.331 **	56.76 ±0.263***
SOD	8.16 ±0.099	5.98±0.075***	3.16±0.262***	8.10±0.136●	7.01±0.175*	7.96 ±0.235**	6.06 ± 0.015***

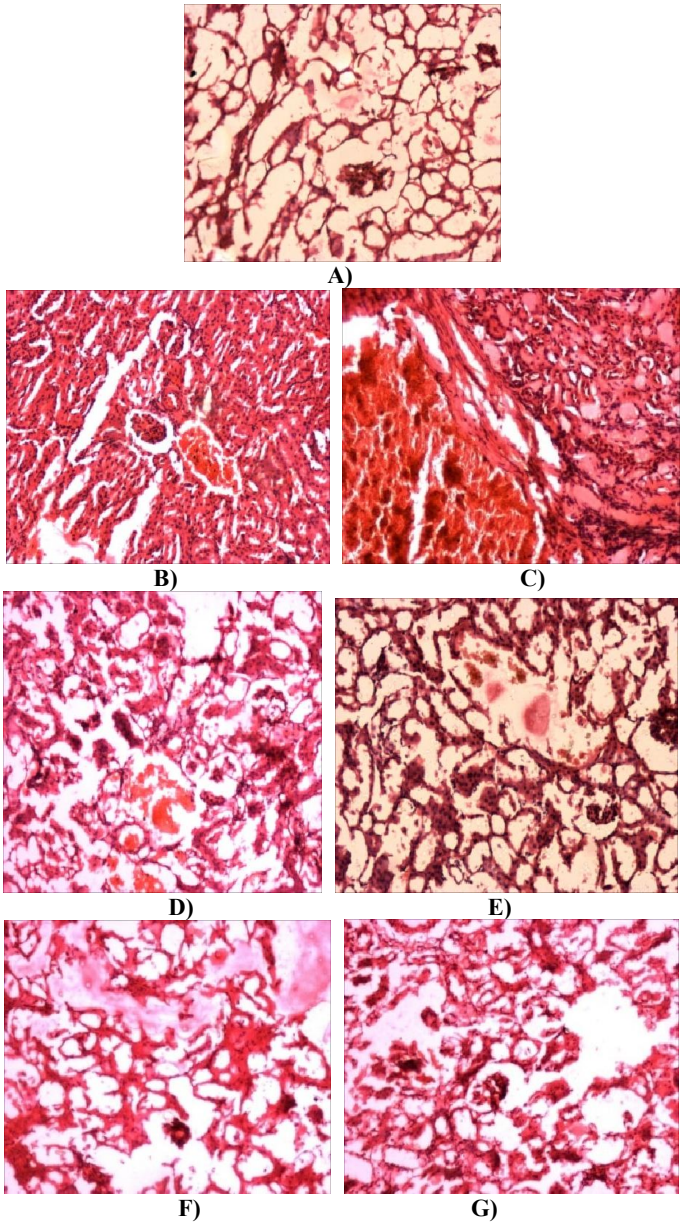

Fig.1. **Histopathological examination of haematoxylin-eosin stained kidney section of normal and experimental rats with magnification ×100. (a)Kidney section of control rat demonstrated the normal cellular structure. (b) Section of the kidney of lindane (for 14 days) alone treated rat showeddegenerative and necrotic changes in proximal convoluted tubules. Note the detachment of necrosed lining cells. (c) Section of the kidney of lindane (for 28 days) alone treated rat showed Marked vascular congestion and tubular dilation. (d) Kidney section of pre-treated rat depictedmild vascular congestion in cortex and medulla with normal structure of glomeruli and medulla.(e) Kidney section of post-treated rat depicted widening of Bowman's space with deposition of proteinaceous mass.(f) Section of the kidney of lindane metabolized rat showed necrotic changes in the proximal convoluted tubules.necrosed lining cells detached in to the luman(g) Kidney section of lindane+curcumin treated rat improved to near normal renal cellular architecture**

Pre and post treatment with curcumin caused a significant increase in renal ($P<0.05$) in SOD and CAT activities suggesting an active protective role by the components present in curcumin in ameliorating free radical- induced damage (Unnikrishnan and Rao 1995). In agreement with our observation Quiles *et al.,* (2002); Ramirez-Tortosa, (2002) reported that curcumin inhibits ROS production which cause oxidative stress. GST plays an important role in the detoxification of toxic electrophiles by conjugating them with glutathione. GST is a potent antioxidant that provides cells with a substantial degree of protection against oxidative stress. In the present study, lindane significantly ($P<0.001$) decreased GST activity in renal tissues **(**Table-1**)**. The decrease in GST activity might be responsible for lindane accumulation in the renal tissues of rat. In support of our findings Padma *et al.,* (2011) has been reported that lindane intoxication decreases the GST level in rat kidney. Spychala (2000) showed a significant decrease in GST in male mice when treated with lindane. In our study, Pre and post treatment with curcumin caused a significant increase in renal ($P<0.05$) GST level in compared to lindane treated rats. This may be due to the Curcumin has been reported as potent scavenger of variety of ROS (Reddy et al.; 1994), exhibiting anti inflammatory activity as well as antioxident properties (Patumraj, *et al*.; 2006, Halim, *et al*.; 2002, Sharma, *et al*.; 2006 and Unnikrishnan and Rao 1995). The phenolic and the methoxy group on the phenyl ring and the 1, 3 _ diketone systems seems to be important structural features that can be potent in scavenging free radicals and the phenolic group with a methoxy at the ortho position is especially effectual for the antioxidant activity (Rao 1994; Priyadarsini *et al*., 2003).

Fig. 1 demonstrated that the rats treated with lindane alone for 14 and 28 days showed degeneration and shrinkage of glomerulus and degeneration of proximal and distal tubules. These alterations in kidney architecture might be due to generation of reactive oxygen species by lindane metabolism which play a deleterious role in causing nephrotoxicity. These findings are similar to previous observations in the same model by Padma *et al.,* (2011) who observed tubular distension and basophilic tubules in kidney. Similarly, exposure of CD and EtOH cause disturbances in histology of rats (Brzóska *et al.,* 2003, 2002).While the Treatment with curcumin to lindane exposed rats reduced the pathomorphological alterations and ameliorated the histomorphology of the hepatic tissue of rats. Renal tissue of rats exposed to lindane showed glomerular mesangial proliferation, proximal tubular cell swelling, glomerular sclerosis, interstial edema (mild), eosinophilic cytoplasm and hydrophic degeneration with satellite lumen. Earlier study reported kidney is a target organ for free redicals which produced by heavy metals and pesticides (Markovich *et al*., 1999). Suter (1983) observed liver and kidney effects in rats fed with lindane showed centrilobular hypertrophy and necrosis, tubular distension and basophilic tubules, respectively. Curcumin administration restored the renal architecture in lindane-exposed rats. In curcumin and lindane co-treated animals only slight degeneration of cells was found.

The result of the present study demonstrate that the lindane induced oxidative damage on the kidneys by enhancing lipid peroxidation and diminishing enzymatic (CAT, GST and SOD) and non-enzymatic (GSH) antioxidant status. Curcumin diminished lindane induced oxidative stress probably through its free radical scavenging, anti-lipid peroxidative and antioxidant activities in the kidneys. Thus, the results of our investigations suggest that curcumin has protective effects on oxidative stress induced by lindane. Curcumin can be a potent antioxidant in the kidneys. These organs are highly prone to oxidative stress against lindane induced toxicity and hence may have useful properties as a natural antioxidant supplement, capable of preventing renal damage caused by oxidative stress and helps in normal functioning of these vital organs.

REFERENCES

Agrawal DP, Sultana, GSD Gupta (1991); Oxidative damage and changes in the glutathione redox system in erythrocytes from rats treated with hexachlorocyclohexane. *Food Chem Toxicol*., 29; 459–462

Ahmed RS, SG Suke, V Seth, A (2008); Chakraborti, A.K.Tripathi and B.D. Banerjee: Protective effects of dietary ginger (*Zingiber officinales* Rosc.) on lindane-induced oxidative stress in rats. *Phytother. Res*., 22; 902-6.

Al-Rejaie SS (2009); Effect of green and black teas on immobilization induced stress in male wistar albino rats. *Int.J.Pharmacol*., 5,137-145.

Ammon HPT, Anazodo, MI, Safayhi, H., Dhawan, BN., Srimal, RC (1992); Curcumin: a potent inhibitor of Leukotriene B4 formation in rat peritoneal polymorphonuclear neutrophils (PMNL). *Planta Med*., 58, 26.

Ananya RS., Subeena, DA., Kumar, DT., Kumar, S.M. Kumar (2005); Oxidative stress and histopathological changes in the heart following oral lindane (g-HCH) administration in rats. *Med.Sci. Monit*., 11, 325-329.

Anderson ME., Powrie, F., Puri., RN., Meister A (1985); Glutathione monoethyl ester: Preparation, uptake by tissues, and conversion to glutathione. *Arch. Biochem. Biophys*., 239: 538-548.

Bandyopadhyay U, Das D, Banerjee RK (1999); Reactive oxygen species: oxidative and pathogenesis. Curr Sci., 77, 658-66.

Banerjee BD, Seth V, Ahmed RS (2001); Pesticides induced oxidative stress: perspectiveness and trends. *Rev. Environ. Health*., 16, 1–40.

Banerjee BD, Seth V, Bhattacharya A, Pasth ST, Chakraborty AK (1999); Biochemical effects of some pesticides on lipid peroxidation and free radical scavengers. *Toxicol. Lett*., 107, 33-47.

Banerjee BD, Zaidi SS, Pasha ST, Rawat DS Koner BC (1997); HCH residues in human milk sample s from Delhi, India. *Bull Environ. Contam .Toxicol*., 59, 403-406.

Bano M, Bhatt DK (2007); Neuroprotective role of a novel combination of certain antioxidants on lindane (g-HCH) induced toxicity in cerebrum in mice. *Res. J. Agric. Biol. Sci*., 3, 664–669.

Barros L A Videla (1994); Acute lindaneintoxication: A study on lindane tissueconcentration and oxidative stress relatrd parameters in liver and erythrocytes. *J. Biochem. Toxicol*., 9, 9-15.

Barros SBM, Simizu K, Junqueira VBC (1991); Liver lipid peroxidation-related parameters after short-term administration of hexazhlorocyclohexane isomers to rats. *Toxicol. Lett*., 56,137–144.

Bhalla P, Agrawal D (1998); Alteration in rat erythrocyte membrane due to hexachlorocyclohexane (technical) exposure. *Hum. Exp. Toxicol*., 17, 638-642.

Bishnoi M, Chopra K, Kulkarni SK (2008); Protective effect of Curcumin, the active principle of turmeric (*Curcuma longa*) in haloperidol-induced orofacial dyskinesia and associated behavioural, biochemical and neurochemical changes in rat brain. *Pharmacology Biochemistry and Behavior*., 88, 511–522.

Bist R, DK Bhatt (2008); The evaluation of effect of alpha-lipoic acid and vitamin E on the lipid peroxidation, gamma-amino butyric acid and serotonin level in the brain of mice (*Mus muscles*) acutely intoxicated with lindane. *Journal of the neurological science*., In press.

Brzoska MM, Moniuszko JJ, Pilat MS (2003); Liver and kidney function and histology in rats exposed to cadmium and ethanol. *Alcohol & alcoholism*., 38, 2-10.

Brzoska MM, Moniuszko JJ, Jurczuk M, Sidorczuk GM (2002); Cadmium turnover and changes of zinc and copper body status of rats continuously exposed to cadmium and ethanol. *Alcohol and Alcoholism*., 37, 213-221.

Budavari S, Neil MJO, Smith A, Heckelman PE (1989); Bioavailability, metabolic effects and safety. *Annu. Rev. Nutr*., 22, 866–867.

Burton GW, Cheesman HN, Ingold KV, Seater TF (1983); Lipid antioxidants and products of lipid peroxidation as potential tumor protective agents. *Biochem. Soc. Trans*., 11, 261–262.

Chadwick RW, Freal JJ (1972); Comparative acceleration of lindane metabolism to chlorophenols by pretreatment of rats with lindane or with DDT and lindane. *Food Cosmet Toxicol* ., 10, 789-795.

Comporti M (1989); Three models of free radical mediated cell injury. *Chem. Biol. Interactions*., 72, 1–56.

El-Demerdash FM, Yousef MI, FM (2009); Radwan Ameliorating effect of curcumin on sodium arsenite-induced oxidative damage and lipid peroxidation in different rat organs. *Food Chem. Toxicol*., 47 , 249-54.

English D, Schell M, Siakotos A, Gabig TG (1986); Reversible activation of the neutrophil superoxide generating system by hexachlorocyclohexane: Correlation with effects on a subcellular superoxide-generating fraction. *J. Immunol*., 137, 283-290.

Fernandez V, Massa L, Quinones KA, Giavarotti S, Almeida VD, Azzalis LA, Junqueira VB, Videla LA (2003); Effects of gamma-hexachlorocyclohexane and L-3,30,5-triiodothyronine on rat liver cytochrome P4502E1-dependent activity and content in relation to microsomal superoxide radical generation. *Biol. Res*., 36, 359–365.

Fidan F, Cigeri H, Sozbilir NB, Kucukkurt I, Yuksel H (2008); The effect of the dose-dependent c-hexachlorocyclohexane (lindane) on blood and tissue antioxidant defense systems, lipid peroxidation and histopathological changes in rats. *J. Anim. Vet. Adv*., 7, 1480–1488.

Fong KL, Mecay PB, Poyer IL, Keele BB, Misra HP (1973); Evidance that peroxidation of lysosomal membranes is initiated by hydroxyl free radicals produced during Flavin enzyme activity. *J. Biol.Chem*., 248, 77–92.

Garcia FAJ, Bayoumi AE, Perez PY (2002); Alterations of the glutathione-redox balance induced by metals in CHO-K1 cells. *Comp Biochem Phys*., 132, 365–373.

Govindarajan VS (1980); Turmeric-chemistry, technology and quality. CRC Cr Rev *In Fd Sci Nut*., 199–301.

Guldur ME, Ozgonul A, Kilic IH, Sogut O, Ozaslan M (2010); Gastroprotective effect of *Cyperus rotundus* extract against gastric mucosal injury induced by ischemia and reperfusion inrats.Int.J.Pharmacol.6,104–110.

Habig WJH, Pabst M, Jakoby WB (1974); Glutathion-s-transferase: The first enzymatic step in mercapturic acid formation. *J. Biol. Chem*., 249, 7130-7139.

Halim E Hussain M (2002); Hypoglycemic, hypolipidemic and antioxidant properties of combination of Curcumin from *CURCUMA LONGA*, Linn, and partially purified product from ABROMA AUGUSTA, Linn, in streptozotocin induced diabetes. *Indian J Clin Biochem*., 17, 33–43.

Hall PF (1994); Testicular steroid synthesis: organisation and regulation. In: Knobil E, Neil JD, editors.. *The physiology of reproduction*. Raven Press, New York, 13, 35-62.

Halliwell B Gutteridge JMC (1989); In: Free radicals in biology and medicine, 2nd edn. Oxford: Clarendon Press.

Hayes JH (1982); Carbamate pesticides In: Pesticides stress induced in man. *J. Biochem*., 7, 436–462.

Hfaiedh N, Claude JM Abdelfettah E (2012); A combination of ascorbic acid and α-tocopherol or a combination of Mg and Zn are both able to reduce the adverse effects of lindane-poisoning on rat brain and liver. *Journal of Trace Elements in Medicine and Biology*.

Iyawe HOT, Onigbinde AO, Aina OO (2006); Effect of chloroquine and ascorbic acid interaction on the oxidative stress status of *Plasmodium berghei* infested mice. *Int. J. Pharmacol.,* 11, 151-169.

Jadon A, Bhadauria M, Shukla S (2007); Protective effect of Terminalia belerica Roxb, and gallic acid against carbon tetrachloride induced damage in albino rats. *Toxicology.*, 12, 165–173.

Jain SK, Rains J Jones K (2006); Effect of curcumin on protein glycosylation, lipid peroxidation, and oxygen radical generation in human red blood cells exposed to high glucose levels. *Free Rad. Biol. Med.*, 41, 92–96.

Joe B Lokesh BR (1994); Role of capsaicin, curcumin and dietary n-3 fatty acids in lowering the generation of reactive oxygen species in rat peritoneal macrophages. *Biochem. Biophys. Acta.*, 1224, 255–263.

Jollow DJ, Mitchell JR, Zampaglione N, Gillette JR (1974); Bromobenzene induced liver necrosis. Protective role of glutathione and evidence for 3,4-bromobenzene oxide as the hepatotoxic metabolite. *Pharmacology.*, 11, 151-169.

Kalender Y, Ogutcu A, Uzunhisarcikli M, Durak D, Acikgoz F (2004); Endosulfan-induced cardiotoxicity and free radical metabolism in rats (2004); The protective effect of vitamin E.*Toxicology*., 202, 227–235.

Kappus H (1987); Oxidative stress in chemical toxicity. *Arch. Toxicol*., 60,144–149.

Kashyap SK (1986); Health surveillance and biological monitoring of pesticide formulators in India. *Toxicol.* Lett., 33, 107–114.

Kerr ME, Bender CM, Monti EJ (1996); An introduction to oxygen free radicals. Heart Lung, 25, 200–209.

Khan SM (2006); Protective effect of black tea extract on the levels of lipidperoxidation and antioxidant enzymes in liver of mice with pesticide-induced liver injury. *Cell Biochem. Funct*., 24, 327-332.

Khanna NM (1999); Turmeric-nature's precious gift. *Curr. Sci*., 76, 1351–1356.

Koner BC, Banerjee BD, Ray A (1998); Organochlorine pesticide induced oxidative stress and immune suppression in rats. *Indian J. Exp. Biol*., 36, 395–398.

kumar Anila KR., Khanum F., Santhanam K. (2006); Amelioration of Hexachlorocyclohexane-Induced Oxidative Stress by Amaranth Leaves in Rats. *Plant Foods for Human Nutrition*., 61, 169-173.

kumar Anila KR., Saritha V., K. Farhath , . Bawa AS (2009) ; Ameliorative effect of ajwain extract on hexachlorocyclohexane-induced lipid peroxidation in rat liver. *Food and Chemical Toxicology*., 47, 279–282.

Kumar NVR, Kuttan R (2003); Modulation of carcinogenic response and antioxidant enzymes of rats administered with 1,2-dimethylhydrazine by picroliv. *Cancer Lett*., 191, 137–143.

Lopez-Aparicio P, del-Hoyo N, Perez-Albarsanz MA (1988); Lindane distribution and phospholipid alterations in rat tissues after administration of linane containing diet. Pest. *Bioch. Physio*., 31, 109–119.

Madesh M Balasubramanian KA (1998); Microtitre plate assay for superoxide dismutase using MTT reduction by superoxide. *Indian J.Biochem. Biophy*., 35,184-188.

Mallick P, Chakrabarti- Mallick J, Guha B, Khuda-Bukhsh AR (2003); Ameliorating effect of microdoses of a potentized homeopathic drug, Arsenicum Album, on arsenic induced toxicity in mice. *BMC Compl. Altern Med*., 3,7.

Markovich D, James KM (1999); Heavy metals mercury, cadmium and chromium inhibit the activity of the mammalian liver and kidney sulfate transporter Sat-1. *Toxicol Appl Pharmacol*., 154, 181-187.

Menzer RE Nelson JO (2000); Water and soil pollutants. In: Klaassen, C.D., Amdur, M.O., Doull, J. (Eds.), *Casarett and Doull's Toxicology*., Macmillan Publishing Co., New York, 825–853.

Moslen MT (1994); Reactive oxygen species in normal physiology, cell injury and phagocytosis. *Adv. Exp. Med. Biol*., 366, 17–27.

Mtgapor, K.J. and J. Fazlina: Effect of various doses of palm vitamin E and tocopherol on factors affecting gastric lesions in rats. *Int. J. Pharmacol*., 2, 98-103 (2006). Mullar L (1986); Consequence of cadmium toxicity in rat hepatocytes: Effect of cadmium on the glutathione peroxidase system. *Toxicol, Lett*., 30, 259–265.

Murphy SD (1986); Toxic effects of pesticides. In: Klaassen, C.D., Amdur, M.O., Doull, J. (Eds.), *Casarett and Doull's Toxicology*., Macmillan Publishing Co., New York, 519–581.

Nandave M, Ojha SK, Joshi S, Kumari S, Arya DS (2007); Cardioprotective effect of *Bacopa monneira* against isoproterenol-induced myocardial necrosis in rats. *Int. J.Pharmacol*,3, 385–392.

Nelson L (1990); Pesticide perturbation of sperm cell function. *Bull. Eviron Contam. Toxicol*., 45 , 876–882.

Nur Azlina MF, Rubaizah K, Muliana MS, Nafeeza MI (2009); Modulation of restraint induced gastric oxidative changes in rats by tocotrienol and tocopherol. *Int. J. Pharmacol*., 5, 58 –64.

Padma VV, Lalitha G, Shirony NP, Baskaran R (2012); Effect of quercetin against lindane induced alterations in the serum and hepatic tissue lipids in wistar rats. *Asian Pacific Journal of Tropical Biomedicine*., 910-915.

Padma VV, Sowmya P, Felix TA, Baskaran R, Poornima P (2011); Protective effect of gallic acid against lindane

induced toxicity in experimental rats. *Food and Chemical Toxicology* 49 ,991–998.

Park EJ, Jeon CH, Ko G, Kim J, Sohn DH (2000); Protective effect of curcumin in rat liver injury induced by carbon tetrachloride. *J Pharm Pharmacol*., 52, 437–440.

Patumraj S, Wongeakin N, Sridulyakul P, Jariyapongskul A, Futrakul N, Bunnag S (2006); Combined effects of curcumin and vitamin C to protect endothelial dysfunction in the iris tissue of STZ-induced diabetic rats. *Clin Hemorheol Microcirc*., 35, 481–9.

PodstawkaV, Grabarezyk M, Kopeeszlezak J (1991); Vitamin E protects human leucocytes against toxic effect of lindane *in vitro*. *Mater. Med. Pol*., 23, 285-289.

Pompella A., Visvikis A, Paolicchi A, Tata VD, Casini AF (2003); "The changing faces of glutathione, a cellular protagonist". *Biochemical Pharmacology*., 66, 1499 -503.

Priyadarsini KI, Maity DK, Naik GH, Kumar MS, Unnikrishnan MK, Satav JG (2003); Role of phenolic O-H and methylene hydrogen on the free radical reactions and antioxidant activity of curcumin. *Free Radic Biol Med*., 35,475–84.

Puatanochokchai R, Morimura K, Wanibuchi H (2006); Alpha-benzene hexachloride exert hormesis in preneoplastic lesion formation of rat hepatocarcinogenesis with the possible role for hepatic detoxifying enzymes. *Cancer Lett*., 240, 102–113.

Pulla RA, Lokesh BR (1992); Studies on spice principles as antioxidants in the inhibition of lipid peroxidation of rat liver microsomes. *Mol Cell Biochem*., 111, 117–124.

Quiles JL, Mesa MD, Ramirez-Tortosa CL, Aguilera CM, Bationo M, Gill A (2002); Curcuminoids as potent inhibitors of lipid peroxidation. *J Pharm Pharmaco*l., 46, 1013–6.

Rajesh MG, Latha MS (2004); Preliminary evaluations of the antihepatotoxic effect of Kamilari, a polyherbal formulation. *J. Ethnopharmacol*. 91, 99-104.

Ramadori G, Moriconi F, Malik I, Dudas J (2008); Physiology and pathophysiology of liver inflammation, damage and repair. *J Physiol Pharmacol*., 59, 107-117.

Ramirez-Tortosa MC (2002); Curcuma longa extract supplementation reduces oxidative stress and attenuates aortic fatty streak development in rabbits. *Arterioscler. Thromb. Vasc. Biol*., 22, 12-25..

Reddy AC Lokesh BR (1994); Studies on the inhibitory effects of curcumin and eugenol on the formation of reactive oxygen species and the oxidation of ferrous iron. *Mol Cell Biochem*., 137, 1–8.

Reed DJ (1990); Glutathione: toxicological implications. Annu. Rev. Pharmacol. Toxicol., 30, 603–631.

Safe SH (1993); Lindane and Hexachlorocyclopentadiene. *Wiley-Intersciene*., 6, 135–139.

Sahoo A, Chainy GBN (1998); Acute hexachlorocyclohexane induced oxidative stress in rat cerebral hemisphere. *Neurochem. Res*., 23,1081-1086.

Samanta L, Sahoo A, Chainy GBN (1999) ; Age related changes in rat testicular oxidative stress parameters by hexchlorocyclohexane. *Arch Toxicol*., 73, 96-107.

Sarhan R., Abd-El-Azim SA, Motawi TMK, Hamdy MA (2007); Protective effect of turmeric, *Ginkgo biloba*, silymarin separately or in combination, on iron-induced oxidative stress and lipid peroxidation in rats. *Int. J. Pharmacol*., 3, 375-384.

Shanmugarajan T.S., Arunsundar M, Somasundaram, I, Krishna kumar, ED. Sivaraman V. Ravi chandiran (2008); Cardioprotective effect of *Ficus hispida* Linn. on cyclophosphamide provoked oxidative myocardial injury in a rat model. *Int. J. Pharmacol*., 4, 78-87.

Sharma S., Kulkarni SK, Chopra K (2006); Curcumin, the active principle of turmeric (Curcuma longa), ameliorates diabetic nephropathy in rats. *Clin Exp Pharmacol Physiol.,* 33, 940–5.

Spychala J 2000; Tumor-promoting functions of adenosine. *Pharmacol. Ther*., 87, 161–173.

Sreejayan Rao MN (1994); Curcuminoids as potent inhibitors of lipid peroxidation. *J Pharm Pharmacol*., 46, 1013-1016.

Srivastava A, Shivanandappa T. (2005);.Hexachlorocyclohexane differentially alters the antioxidant status of the brain regions in rat.Toxicol., 214: 123–130.

Suter P (1983); Three months toxicity study in rats with lindane. *Toxicology*., 11, 42–51.

Tappel AL (1974); Lipid peroxidation damage to cell components. *Fed. Proc*., 32, 1870–1874.

Unnikrishnan MK., Rao MN (1995); Inhibition of nitrite induced oxidation of hemoglobin by curcuminoids. *Pharmazie*., 50: 490–492.

Videla, L.A., Barros SB, Janqueria VB 1990; Lindane induced liver oxidative stress. *Free Raddic. Biol .Med*., 9, 69–179.

Vijayavel K., Anbuselvam C, Balasubramanian MP (2006); Free radical scavenging activity of the marine mangrove *Rhizophora Apiculata* bark extract with reference to naphthalene induced mitochondrial dysfunction. *Chem. Biol. Interact*., 163, 170-175.

Wauchope RD., Buttler TM., Hornsby AG., Augustijn-Beckers PW, Burt JP. (1992); The SCS/ARS/CES pesticide properties database for environmental decision making. *Rev. Environ. Contam. Toxicol*., 123, 1–155.

Xavier R., Rekha K.., Bairy KL (2004); Health perspective of pesticide exposure anddietary management. *Malays. J. Nutr*., 10, 39–51.

Yavuz T, Delibao N., Yaldaram B, Altuntas I, Candar O, Cora A., Karahan N., Ibrisim E. Kutsal A. (2005); Vascular wall damage in rats induced by organophosphorus insecticide ethidation, *Toxicol. Lett*., 155, 59–64.

****