academicJournals

Vol. 10(38), pp. 810-816, 15 October, 2016
DOI: 10.5897/AJPP2016.4673
Article Number: C30DCF961125
ISSN 1996-0816

http://www.academicjournals.org/AJPP

African Journal of Pharmacy and Pharmacology

Full Length Research Paper

In vivo antioxidant and lipid peroxidation effect of various extracts from aerial parts of *Chomelia asiatica* (Linn) in rat fed with high fat diet

Abdul Hameed Thayyil[2], A. Kottai Muthu[1]* and Mohammed Ibrahim[2]

[1]Department of Pharmacy, Annamalai University, Annamalai Nagar-608 002, India.
[2]Nizam Institute of Pharmacy and Research centre, Near Ramoji Film City, Deshmukhi, Hyderabad, A.P., India.

Received 12 September, 2016: Accepted 30 September, 2016

The present investigation was to examine the *in vivo* antioxidant activity and lipid peroxidation activities of different extracts of aerial parts of *Chomelia asiatica* (Linn). High fat diet rats demonstrated fundamentally decreased the levels of tissues enzymatic antioxidant and non-enzymatic antioxidant (Glutathione). The level of thiobarbuturic acid reactive substance (TBARS) is reduced in high-fat diet (HFD) rats when compared and control group. Administration of ethyl acetate extract of *Chomelia asiatica* in high fat diet rats were indicated altogether (p<0.001) increased the levels of antioxidant enzymes, for example, superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx), glutathione reductase (GR) and level of non enzymatic antioxidant glutathione (GSH) when contrasted and HFD rats (Group II). The ethyl acetate extract of *C. asiatica* in high fat diet rats were discovered lowered the concentration of TBARS when contrasted and HFD rats. In comparison of all the three extracts treated group with standard group, the ethyl acetate extract of *C. asiatica* showed significant (p<0.001) result than that of other groups. Taking into account the outcomes, we concluded that the ethyl acetate extract of *C. asiatica* is a significant source of antioxidant, which may be useful in keeping the advancement of different oxidative stresses.

Key words: *Chomelia asiatica*, *in vivo* antioxidant, lipid peroxidation, rats.

INTRODUCTION

Oxidation is essential in many living organisms for the production of energy to fuel biological processes. However, the uncontrolled production of oxygen derived free radicals is involved in the onset of many diseases such as atherosclerosis, rheumatoid arthritis, and cancer as well as in degenerative processes associated with aging (Halliwell et al 1984). Almost all organisms are well protected against free radical damage by enzymes such as superoxide dismutase and catalase, or compounds such as ascorbic acid, tocopherols and glutathione (Mau et al., 2002). When the mechanism of antioxidant protection becomes unbalanced by factors such as aging, deterioration of physiological functions may occur resulting in diseases and accelerated aging (Israel, 2012).

*Corresponding author. E-mail: arthik03@yahoo.com. Tel: 91-9443171712.

However, the antioxidants present in human diet are of great interest as possible protective agents to help the human bodies reduce oxidative damage. Antioxidants are often used in oils and fatty foods to retard their autoxidation. Synthetic antioxidants, such as butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA), have restricted use in foods as they are suspected to be carcinogenic. Therefore, the importance of search for natural antioxidants has greatly increased in the recent years (Jayaprakasha et al., 2003). Ethnomedical literature contains a large number of plants that can be used against diseases, in which reactive oxygen species and free radical play important role. There is a plethora of plants that have been found to possess strong antioxidant activity (Badami et al., 2003). Recent reports indicate that there is an inverse relationship between the dietary intake of antioxidant-rich foods and the incidence of human diseases (Halliwell, 1997). So, many researchers have focused on natural antioxidants and in the plant kingdom numerous crude extracts and pure natural compounds were previously reported to have antioxidant properties. However, no data are available in the literature on the antioxidant activity of aerial parts of *C. asiatica*. Therefore we undertook the present investigation to examine the *in vivo* antioxidant and lipid peroxidation activities of different extract of aerial parts of *C. asiatica* (Linn) in rat fed with high fat diet.

MATERIALS AND METHODS

Chemicals and reagents

Unless stated, general chemicals and reagents were purchased from Sigma, Cadila. Fisher chemicals were of analytical grade or equivalent.

The plant materials (Study species)

C. asiatica (Linn) belongs to the family Rubiaceae, commonly known as *Tharani in tami*: *Kuppipoovu, Tharana* in makayalam. *Chomelia Gaertn* is a genus of about 370 species distributed in tropical and subtropical Africa, Asia, Madagascar and pacific islands*. Chomelia asiatica* (Linn) is a common species which occurs in india, Srilanka and China. The leaves or powder extracts of *C. asiatica* are used as antimicrobial activities (Jayasinghe et al., 2002). It had been accounted for pain relieving and mitigating activities (Amutha et al., 2012). The parts of *Tarenna asiatica* (Rubiaceae) plants are customarily used to advance suppuration (Anonymous, 1976), as anthelmintic (Ramarao and Henry, 1996) and antiulcer operator (Rao et al., 2006). The phytochemical constituents of it are accounted for to be antiseptic (Vinothkumar et al., 2011), injury healing (Anjanadevi and Menaga, 2013) and antioxidant (Ramabarathi et al., 2014). Moreover, the extract of shoots, leaves and fruits are purportedly dynamic against *Mycobacter phlei* (Rajakaruna et al., 2002).

Collection and identification of plant materials

The aerial parts of *C. asiatica* (Linn), were collected form Shencottai, Tirunelveli District, Tamil Nadu, India. Taxonomic identification (Plant deposition no is Au/CA/810) was made from Botanical Survey of Medical Plants Unit Siddha, Government of India. Palayamkottai, Tirunelveli. The aerial parts of *C. asiatica* (Linn)*,* were dried under shade, separated, pulverized by a mechanical processor and went through a 40 mesh sieve.

Preparation of extracts

The above powered materials were successively extracted with petroleum ether (40-60°C) by hot continuous percolation technique in Soxhlet apparatus (Harborne JB, 1984) for one day. Then the marc was subjected to ethyl acetate (76-78°C) for one day and then mark was subjected to methanol for one day. The extracts were extracted by utilizing a rotary evaporator and subjected to freeze drying in a lyophilizer till dry powder was obtained.

Animals and treatment

Male Wister rats of 16 to 19 weeks age, weighing 150 to 175 g were obtained from the Central Animal House, Nizam Institute of Pharmacy and Research Centre, Near Ramoji Film City, Deshmukhi, Hyderabad, Telugana, India. The animals were kept in cages, 2 per confine, with 12:12 hr light and dim cycle at 25±2°C. The animals were maintained on their separate diets and water *ad libitum.* Animal Ethical Committee's clearance (Approval number is NIPRC/IAEC/Ph.D/2015/01) was obtained for the study.

Experimental design

Rats were divided into following 6 groups of 6 rats each:

I group: Standard chow pellet
II group: HF Diet
III group: HF diet + petroleum ether extract of *Chomelia asiatica* (Linn) (200 mg/kg B.wt)
IV group: HF diet + Ethyl acetate extract of *Chomelia asiatica* (Linn) (200 mg/kg B.wt)
V group: HF diet + Methanolic extract of *Chomelia asiatica* (Linn) (200 mg/kg B.wt)
VI group: HF diet + Standard drug atorvastatin (1.2 mg/kg B.wt)

Animal diet

The compositions of the two diets were as follows (Kottai et al., 2005):

Control diet: Wheat flour 22.5%, simmered bengal gram powder 60%, skimmed milk powder 5%, casein 4%, refined oil 4%, salt blend with starch 4% and vitamin and choline blend 0.5%.
High fat diet: Wheat flour 20.5%, broiled bengal gram 52.6%, skimmed milk powder 5%, casein 4%, refined oil 4%, coconut oil 9%, salt blend with starch 4% and vitamin and choline blend 0.5%, cholesterol 0.4%.

Assessment of *in vivo* antioxidant and lipid peroxidation

Rats of III, IV and V groups were orally fed with the different extracts of *C. asiatica* (Linn) and rats of VI group were fed with standard drug atorvastatin. Both the extract and atorvastatin were suspended in 2% tween 80 (Waynforth BH et al., 1980) separately and fed to the respective rats by oral intubation. At the end of sixty three days all the animals were sacrificed by cervical dislocation

Table 1. Effect of various extracts of aerial parts of *Chomelia asiatica* (Linn) on tissues TBARS in HF diet rats.

Groups	Thio barbituric acid reactive substances (TBARS)		
	Liver	Heart	Aorta
I	$25.50 \pm 0.158^{b}*$	$44.14 \pm 0.121^{b}*$	$16.47 \pm 0.104^{b}**$
II	$77.39 \pm 0.104^{a}*$	$87.37 \pm 0.129^{a}*$	$66.63 \pm 0.702^{a}*$
III	$68.42 \pm 0.167^{a}**,^{b}**$	$78.49 \pm 0.164^{a}**,^{b}*$	$55.30 \pm 0.114^{a}**,^{b}*$
IV	$34.15 \pm 0.078^{a}**,^{b}*$	$47.21 \pm 0.078^{a}**,^{b}*$	$26..32 \pm 0.141^{a}**,^{b}*$
V	$40.27 \pm 0.118^{a}*,^{b}*$	$58.34 \pm 0.069^{a}*,^{b}*$	$30.46 \pm 0.07^{a}*,^{b}*$
VI	$28.33 \pm 0.143^{a}*,^{b}*$	$43.35 \pm 0.152^{a}*,^{b}*$	$18.57 \pm 0.135^{a}*,^{b}*$

Values are mean ± SE of 6 rats. *P* values: * < 0.001, ** < 0.05; a → groups II, III, IV, V and VI compared with group I (control). b → groups I, III, IV, V and VI compared with group II (High fat diet). I group: standard chow pellet. (Control); II group: High Fat Diet. III group: High fat diet + Petroleum ether extract of *C. asiatica* (200mg/kg B.wt); IV group: High fat diet + Ethyl acetate extract of *C.asiatica* (200 mg/kg B.wt);V group: High fat diet + Methanolic extract of *C. asiatica* (200mg/kg B.wt); Group VI: High fat diet + standard drug atorvastatin (1.2 mg/kg B.wt).

Table 2. Effect of various extract of aerial parts of *C. asiatica* (Linn)on tissues conjugated diene in HF diet rats.

Groups	Conjugated diene (μmoles /g tissue)		
	Liver	Heart	Aorta
I	$175.18\pm0.07^{b}*$	$162.29\pm0.11^{b}*$	$472.52\pm0.36^{b}*$
II	$291.23\pm0.37^{a}*$	$271.32\pm0.34^{a}*$	$744.65\pm0.25^{a}*$
III	$277.85\pm0.22^{a}**,^{b}*$	$250.92\pm0.297^{a}*,^{b}*$	$710.48\pm0.18^{a}*,^{b}*$
IV	$191.86\pm0.21^{a}*,^{b}*$	$179.01\pm0.32^{a}**,^{b}*$	$486.83\pm0.25^{a}**,^{b}*$
V	$222.30\pm0.14^{a}*,^{b}**$	$224.34\pm0.13^{a}*,^{b}*$	$646.04\pm0.17^{a}**,^{b}*$
VI	$199.77\pm0.16^{aNS,b}*$	$172.09\pm0.16^{a}*,^{b}*$	$471.03\pm0.34^{a}**,^{b}*$

Values are mean ± SE of 6 rats. *P* values: * < 0.001, ** < 0.05; a → groups II, III, IV, V and VI compared with group I (control) . b →groups I, III, IV, V and VI compared with group II (High fat diet). Group I-VI details are same as in Table 1.

after overnight fasting. Liver, heart and aorta were cleared of sticking fat, weighed precisely and utilized for the preparation of homogenate. Animals were given enough care as per the Animal Ethical Committee's recommendations. Portion of the tissues from liver, heart and aorta were marked, weighed and homogenized with methanol (3 volumes). The lipid extract obtained by the method of Folch et al. (1957). It was used for the estimation of thiobarbituric acid reactive substances (TBARS) (Nichans et al., 1968). Another portion of the tissues was homogenized with phosphate buffer pH 7.4(0.01 M) and used for the estimation of glutathione (GSH) (Ellman et al., 1959) , superoxide dismutase (SOD) (Kakkar et al., 1984), catalase (CAT) (Sinha et al., 1972), glutathione peroxidase (GPx) (Rotruck et al., 1973), Glutathione Reductase (GR) (Mavis et al., 1968) and glutathione-S-transferase (GST) (Habig et al., 1974).

Statistical analysis

Results were communicated as mean ± SE of 6 rats in every group. The statistical significance between the groups was analyzed by utilizing one way analysis of variance (ANOVA), followed by Dunnet's multiple correlation test. Significance level was fixed at 0.05.

RESULTS

As shown in Tables 1 and 2. The TBARS and conjugated dienes levels were increment in liver, heart and aorta in II group rats are a clear reasonable sign of excessive formation of free radical and initiation of lipid peroxidation. The significantly ($p<0.001$) decreased the levels of TBARS and conjugated dienes, in rats regulated with ethyl acetate extract of *C. asiatica* along with HF diet (IV group) when contrasted to HF diet rats (II group).

Tables 3 and 4 demonstrates that the effect of various extracts of *C. asiatica* on tissues SOD and CAT enzyme levels in HF diet rats. The activities of SOD and CAT in the tissue like liver, heart and aorta were significantly ($P<0.001$) brought down in rats fed with high fat diet (II group) than that of control group .After treatment of ethyl acetate extract of *C. asiatica* along with HF diet significantly expands the activities of SOD and CAT in tissues of rats when compared with other extracts.

The activities of tissues glutathione reductase (GR), glutathione peroxidase (GPx) and glutathione-S-transferese in HF diet rats were presented in Tables 5 to 7. Tissues glutathione peroxidase and glutathione reductase levels were significantly ($p<0.001$) lowered in rats sustained with HF diet (II group) when compared with the control rats (I group). Administration of ethyl acetate of *C. asiatica* alongside the HF diet significantly

Table 3. Effect of various extracts aerial parts of *C. asiatica* (Linn) on tissues SOD in HF diet rats.

Groups	Superoxide dismutase		
	Liver	Heart	Aorta
I	3.65 ± 0.06^{b}*	1.82 ±0.03^{b}*	2.88 ±0.02^{b}*
II	1.75 ±0.02^{a}*	0.78 ±0.03^{a}*	1.49 ±0.02^{a}*
III	2.14 ± 0.02^{a}**,^{b}**	1.03 ± 0.02 ^{a}**,^{b}**	1.71 ± 0.02^{a}**,^{b}*
IV	3.17 ± 0.04^{a}**,^{b}*	1.61 ± 0.02^{a}**,^{b}*	2.63 ± 0.04^{a}**,^{b}**
V	2.50 ± 0.06^{a}*,^{b}*	1.22 ± 0.01^{a}*,^{b}*	2.44 ± 0.02^{a}*,^{b}*
VI	3.73 ± 0.03^{a}*,^{b}*	1.85 ± 0.02^{a}*,^{b}*	2.80 ± 0.02^{a}*,^{b}*

Values are mean ± SE of 6 rats. *P* values: * < 0.001, ** < 0.05; a → groups II, III, IV, V and VI compared with group I (control). b →groups I, III, IV, V and VI compared with group II (High fat diet). Group I-VI details are same as in Table 1.

Table 4. Effect of various s from aerial parts of *C. asiatica* (Linn) on tissues CAT in HF diet rats.

Groups	Catalase		
	Liver	Heart	Aorta
I	29.19 ± 0.10^{b}*	46.28 ± 0.10^{b}*	31.44 ± 0.04^{b}*
II	15.17 ± 0.06^{a}*	30.26 ± 0.05^{a}*	20.43 ± 0.05^{a}*
III	17.25 ± 0.05^{a}**,^{b}**	32.18 ± 0.06^{a}**,^{b}*	22.27 ± 0.07^{a}**,^{b}**
IV	26.13 ± 0.04^{a}**,^{b}*	45.19 ± 0.06^{a}**,^{b}*	29.27 ± 0.06^{a}**,^{b}*
V	20.72 ± 0.23^{a}*,^{b}*	40.38± 0.13^{a}*,^{b}*	24.36 ± 0.10^{a}*,^{b}*
VI	30.30 ± 0.10^{a}*,^{b}*	48.38 ± 0.10^{a}*,^{b}*	30.23 ± 0.03^{a}*,^{b}*

Values are mean ± SE of 6 rats. *P* values: * < 0.001, ** < 0.05; a → groups II, III, IV, V and VI compared with group I (control).b →groups I, III, IV, V and VI compared with group II (High fat diet). Group I-VI details are same as in Table 1.

Table 5. Effect of various extracts of aerial parts of *C. asiatica* (Linn) on tissues glutathione peroxidase in HF diet rats.

Groups	Glutathione peroxidase		
	Liver	Heart	Aorta
I	8.81 ± 0.04^{b}*	15.67 ± 0.02^{b}*	14.17 ± 0.03^{b}*
II	5.19 ± 0.04^{a}*	7.26 ± 0.03^{a}*	6.80 ± 0.03^{a}*
III	5.87 ± 0.04^{a}**,^{b}**	8.16 ± 0.03^{a}**,^{b}**	7.20 ± 0.04^{a}**,^{b}**
IV	8.14 ± 0.04^{a}**,^{b}*	14.12 ± 0.02^{a}**,^{b}*	13.82 ± 0.03^{a}**,^{b}*
V	6.91 ± 0.05^{a}*,^{b}*	12.19 ± 0.04^{a}*,^{b}*	10.40± 0.04^{a}*,^{b}*
VI	8.43 ± 0.06^{a}*,^{b}*	16.19 ± 0.04^{a}*,^{b}*	15.75 ± 0.04^{a}*,^{b}*

Values are mean ± SE of 6 rats. *P* values: * < 0.001, ** < 0.05; a → groups II, III, IV, V and VI compared with group I (control).b →groups I, III, IV, V and VI compared with group II (High fat diet). Group I-VI details are same as in Table 1.

Table 6. Effect of various extracts of aerial parts of *C. asiatica* (Linn) on tissues Glutathione Reductase in HF diet rats.

Groups	Glutathione reductase		
	Liver	Heart	Aorta
I	1.55 ± 0.04^{b}*	2.67 ± 0.05^{b}*	1.70 ± 0.02^{b}*
II	0.63 ± 0.12^{a}*	1.16 ± 0.03^{a}*	0.80 ± 0.01^{a}*
III	0.81 ± 0.02^{a}**,^{b}**	1.75 ± 0.03^{a}**,^{b}**	0.94 ± 0.01^{a}**,^{b}**
IV	1.37 ± 0.02^{a}**,^{b}*	2.58 ± 0.04^{a}**,^{b}*	1.67 ± 0.02^{a}**,^{b}*
V	1.10 ± 0.02^{a}*,^{b}*	1.98 ± 0.03^{a}*,^{b}*	1.30 ± 0.19^{a}*,^{b}*
VI	1.62 ± 0.03^{a}*,^{b}*	2.80 ± 0.02^{a}*,^{b}*	1.77 ± 0.01^{a}*,^{b}*

Values are mean ± SE of 6 rats. *P* values: * < 0.001, ** < 0.05; a → groups II, III, IV, V and VI compared with group I (control).b →groups I, III, IV, V and VI compared with group II (High fat diet). Group I-VI details are same as in Table 1.

Table 7. Effect of various extracts of aerial parts of *C. asiatica* (Linn) on tissues Glutathione S-Transferase in HF diet rats.

Groups	Glutathione – S – transferase (GST)		
	Liver	Heart	Aorta
I	25.38±0.04^{b}*	19.29±0.04^{b}*	17.23±0.04^{b}*
II	10.24±0.03^{a}*	8.65±0.02^{a}*	7.42±0.05^{a}*
III	12.17±0.02^{a}*, ^{b}*	9.15±0.02^{a}*, ^{b}*	7.95±0.05^{a}*,^{b}*
IV	19.45±0.02^{a}*, ^{b}*	16.33±0.03^{a}*, ^{b}*	14.17±0.02^{a}*, ^{b}*
V	13.20±0.04^{a}*, ^{b}*	11.44±0.03^{a}*, ^{b}*	9.32±0.05^{a}*, ^{b}*
VI	21.17±0.03^{a}*, ^{b}*	17.39±0.03^{a}*, ^{b}*	15.28±0.07^{a}*, ^{b}*

Values are mean ± SE of 6 rats. *P* values: * < 0.001, ** < 0.05; a → groups II, III, IV, V and VI compared with group I (control).b →groups I, III, IV, V and VI compared with group II (High fat diet). Group I-VI details are same as in Table 1.

Table 8. Effect of various extracts of aerial parts of *Chomelia asiatica* (Linn) on tissues glutathione in HF diet rats.

Groups	Glutathione		
	Liver	Heart	Aorta
I	4.33 ± 0.04^{b}**	7.68 ± 0.04 ^{b}*	5.69 ± 0.07^{b}*
II	1.71 ± 0.03^{a}**	4.13 ± 0.03^{a}*	2.73 ± 0.05^{a}*
III	2.00 ± 0.03^{a}**,^{b}**	4.27 ± 0.03^{a}**,^{b}**	3.16 ± 0.03^{a}**,^{b}**
IV	4.02 ± 0.04^{a}**,^{b}**	6.78 ± 0.02^{a}**,^{b}*	5.20 ± 0.04^{a}**,^{b}*
V	3.11 ± 0.02^{b}*	5.14 ± 0.02^{b}*	3.85 ± 0.05^{b}*
VI	4.42 ± 0.^{b}*	7.83 ± 0.04^{b}*	5.66 ± 0.05^{b}*

Values are mean ± SE of 6 rats. *P* values: * < 0.001, ** < 0.05; a → groups II, III, IV, V and VI compared with group I (control).b →groups I, III, IV, V and VI compared with group II (High fat diet). Group I-VI details are same as in Table 1.

(p<0.001) increase the levels of glutathione peroxidase, glutathione reductase and glutathione-S-transferese in every one of the tissues as contrasted and HF diet rats. A standard drug atorvastatin regulated rats likewise demonstrated raised level of glutathione peroxidase, glutathione reductase and glutathione-S-transferese.

As shown in Table 8. The significant (p<0.001) fall in the levels of tissues glutathione (GSH) were seen in high fat diet rats (II group) when compared with the control rats (I group).Administration of ethyl acetate extract of *C. asiatica* alongside HF diet rats significantly (p<0.001) increase the levels of glutathione when contrasted and HF diet rats (II group).Similarly the ethyl acetate extract of *C. asiatica* was indicated significant result than that of other two treated groups.

DISCUSSION

The elevated levels of TBARS and conjugated denies were observed in aorta, heart and liver of rats fed with high fat diet (group II). The high fat diet is known to induce oxidative stress in the cells by producing reactive oxygen species (ROS) (Khan et al., 2004). This results in increased lipid peroxidation leading to elevated concentration of TBARS and conjugated dienes (Boccio et al., 1990). The significant decrease in the level of TBARS and conjugated dienes in rats administered with ethyl acetate extract of *C. asiatica* .This effect may be due to phytoconstituents, flavonoids present in the *c. asiatica*.

The activities of SOD and CAT in the tissue like liver, heart and aorta significantly (P<0.001) lowered in rats fed with high fat diet (group II). High fat diet can cause the formation of toxic intermediates that can inhibit the activity of antioxidant enzymes (Thampi et al., 1991) and the accumulation of O_2^- and H_2O_2 which in turn forms hydroxyl radicals (Batra et al., 1989). Catalase decomposes hydrogen peroxide and helps to protect the tissues from highly reactive hydroxyl radicals. Administration of with ethyl acetate extract of *C. asiatica* along with high fat diet significantly increased the activities of SOD and CAT in tissues of rats when compared to other extracts treated groups.

The results indicated that the concentration of glutathione peroxidase (GPX), glutathione reductase and glutathione-s transferase significantly decreased in tissues (aorta, heart and liver) of rats fed with high fat diet. High fat diet decreased the ratio of oxidized glutathione/reduced glutathione in tissue (De La Cruz et al., 2000). Administration of acetate extract of *C. asiatica* along with the high fat diet increased the activities of

glutathione peroxidase, glutathione reductase and glutathione S-transferase in all the tissues. It might be due to help to propagation of biological membranes found to be associated with increase in the activities of GPX. Glutathione peroxidase (GPX) mainly detoxifies H_2O_2 (Izawa et al., 1996) and is involved in the reduction of a variety of hydroperoxides such as phospholipid hydroperoxides, fatty acid hydroperoxides.

Glutathione (GSH), a tripeptide which is present in all the cells is an important antioxidant (Chance, 1992). GSH also functions as free radical scavenger in the repair of radical caused biological damage (Meister, 1984). The reduced levels may be an attempt by the tissue to counteract the increased formation of lipid peroxides that are handled by antioxidant enzymes such as Glutathione peroxidase which scavenges H_2O_2 utilizing GSH as substrate (Rajashree et al., 1998). Increase in glutathione concentration in *C. asiatica* extract treated rats with high fat diet might be due to the increase in the activity of the enzyme glutathione reductase which catalyses the conversion of oxidized glutathione to reduced glutathione in liver (or) might be due to enhanced synthesis of GSH (Sethupathy et al., 2002).

Conclusion

The results of the above study clearly indicated that the ethyl acetate extract of aerial parts *C. asiatica* had significant *in vivo* antioxidant and lipid peroxidation action. The phytoconstituents might be responsible for the inhibition of lipid peroxidation and enhance the antioxidant activities of ethyl acetate extract of *C. asiatica*.

Conflicts of Interests

The authors have not declared any conflict of interests**.**

ACKNOWLEDGEMENT

The authors are thankful to the Annamalai University, Annamalai nagar, Chidambaram, India and Nizam Institute of Pharmacy and Research Centre, Near Ramoji Film City, Deshmukhi, Hyderabad, Telugana, India for providing laboratory and technical support for the present investigation.

REFERENCES

Amutha D, Shanthi S, Mariappan V (2012). Antiinflammatory effect of Tarenna asiatica in carrageenan induced lung inflammation. Int. J. Pharm. Pharmaceut. Sci. 4:344-347.

Anjanadevi N, Menaga S (2013). Wound healing potential of Tarenna asiatica leaves. J. Theor. Exp. Biol. 10:75-80.

Anonymous (1976). The wealth of India: Raw materials. New Delhi, Council of Scientific and Industrial Research. pp. 130-131.

Badami S, Gupta MK, Suresh B (2003). Antioxidant activity of ethanolic extract of *Striga orobanchioides*, J. Ethnopharmacol. 85:227-230.

Batra S, Singh SP, Srivasta VML (1989). Xanthine oxidase, Superoxide dismutase, Catalase and lipid peroxidation in mastomys nataensis effect of dipentalonema viteae infection, Indian J. Exp. Biol. 27:1067.

Boccio GD, Lapenna D, Porreca E, Pennelli A, Savini F, Feliciani P, Ricci G, Cuccurullo F (1990). Aoertic antioxidant defense mechanisms: time related changes in cholesterol fed rabbits. Atherosclerosis pp. 81-127.

Chance B, Greenstein DS (1992). The mechanism of Catalase actions-steady state analysis. Arch. Biochem. Biophys. 37:301-339.

De La Cruz JP, Quintero L, Villalobos MA, Sanchez de.la Cuesta F (2000). Lipid peroxidation and glutathione system in hyperlipedemic rabbits influence of olive oil administration. Biochem. Biophys. Acta.1485:36.

Ellman GL (1959). Tissue sulfhydroyl groups. Arch. Biochem. Biophy. 82:70.

Folch J, Lees M, Sloane GH (1957). A simple method for the isolation and purification of total lipids from animals tissues. J. Biol. Chem. 226:497.

Habig WH, Pabst MJ, Jakoby WB (1974). Glutathione – S – transferase, the first enzymatic step in mercaptouric acid formation. J. Biol. Chem. 249:7130- 7139.

Halliwell B, Gutteridge JMC (1984). Lipid peroxidation, oxygen radicals, cell damage, and antioxidant therapy, The Lancet, 323:1396-1397.

Halliwell B (1997). Advances in pharmacology, Academic Press 38:3-17.

Harborne JB (1984). Phytochemical methods 11 Edn.In Chapman &, Hall. New York. pp. 4-5.

Izawa S, Inoue Y, Kimura A (1996). Importance of Catalase in the adaptive response to hydrogen peroxide analysis of a catalasaemic Saccharomyces Cerevisae. Biochem. J. 320: 61-67.

Jayaprakasha GK, Selvi T, Sakariah KK (2003). Antibacterial and antioxidant activities of grape (Vitis vinifera) seed extract. Food Res Int., 36: 117–122.

Jayasinghe ULB, Jayasooriya CP, Bandara BMR, Ekanayake SP, Merlini Assante LG (2002). Antimicrobial activity of Sri Lankan Rubiaceae and Meliaceae. Fitoterapia. 73:424-427.

Kakkar P, Das B, Visvanathan PN (1984). A modified spectrophotometric assay of SOD, Indian J. Biochem. Biophys. 21:130-132.

Khan SA, Lee K, Minhas KM, Gonzalez DR, Raju SV, Tejani AD (2004). Neuronal nitric oxide synthase negatively regulates xanthine oxidoreductase inhibition of cardiac excitation-contraction coupling. Proc. Natl. Acad. Sci. USA. 101:15944.

Kottai Muthu A, Sethupathy S, Manavalan R, Karar PK (2005). Hypolipidemic effect of methanolic extract of *Dolichos biflorus* Linn in high fat diet fed rats. Ind. J. Exp. Biol. 43:522-525.

Mau JL, Lin HC, Song SF (2002). Antioxidant properties of several specialty mushrooms. Food Res. Int, 35: 519-526.

Mavis RD, Stellwagen E (1968). Purification and Subunit Structure of Glutathione Reductase from Bakers' Yeast. J. Biol. Chem. 243:809-814.

Meister A (1984). New aspects of glutathione biochemistry and transport selective alterations of glutathione metabolism. Nutr. Rev. 42:397.

Nichans WH, Samulelson B (1968). Formation of malondialdehyde from phospholipid arachidonate during microsomal lipid peroxidation. Euro. J. Biochem. 6: 126-130.

Rajakaruna N, Harris CS, Towers GHN (2002). Antimicrobial activity of plants collected from serpentine outcrops in Sri Lanka. Pharmaceut. Biol. 40:235-244.

Rajashree GR, Rajmohan J, Augusti KT(1998). Antiperoxide effect of garlic Protein in alcohol fed rats. Ind. J. Exp. Biol. 36:60.

Ramabharathi V, Apparao AVN, Rajith G (2014). Phytochemical investigation and evaluation of antibacterial and antioxidant activities of leaf-bud exudate of *Tarenna asiatica* (L.) Kuntze ex K. Schum. Indian J. Nat. Prod. Resour. 5:48-51.

Ramarao N, Henry AN (1996). The ethnobotany of Eastern Ghats in Andhra Pradesh, India. Calcutta. Botanical Survey of India.

Rao DM, Rao UVUB, Sudharshanam D (2006). Ethno-medico-botanical studies from Rayalaseema region of southern Eastern Ghats, Andhra

Pradesh, India. Ethnobotanical Leaflets. 10:198-207.
Rotruck JT, Pope AL, Ganther HE, Swanson AB, Hatman DG, Hoekstra WG (1973). Selenium; Biochemical roles as a component of glutathione peroxidise. Science 179:588.
Sethupathy S, Elanchezhiyan C, Vasudevan K, Rajagopal G (2002). Antiatherogenic effect of taurine in high fat fed rats. Ind. J. Exp. Biol. 40: 1169.
Sinha AK (1972). Colorimetric assay of catalase. Anal. Biochem. 47:389.
Thampi HBS, Manoj G, Leelamma S, Menon VG (1991). Dietary fibre and lipid peroxidation: Effects of dietary fibre on levels of lipids and lipid peroxides in high fat diet, Ind. J. Exp. Biol. 29:563.
Vinothkumar D, Murugavelh S, Prabhavathy AK (2011). Phytosociological and ethnobotanical studies of sacred groves in Pudukottai district, Tamil Nadu, India. Asian J. Exp. Biol. Sci. 2: 306-315.
Waynforth BH (1980). Injection techniques. Experimental and surgical techniques in the rats, Academic Press, London. 3 p.