The Pharma Innovation Journal 2017; 6(12): 149-156

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating 2017: 5.03
TPI 2017; 6(12): 149-156

www.thepharmajournal.com
Received: 06-10-2017
Accepted: 07-11-2017

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Evaluation of effect of acetaminophen and *Eclipta alba*. L extract on biochemical parameters on liver of Rattus rattus

Sumira Aziz and Mohd Ashraf Ganaie

Abstract
The present study of the preliminary phyto chemical qualitative examination of acetaminophen (paracetamol) and aqueous extracts of *Eclipta alba L.* shows presence of different biochemical parameters, The various biochemical parameters namely SGOT, SGPT SALP serum, bilirubin and total protein were monitored in this study, because of their diagnostic significance and role in providing information concerning biochemical changes caused by acetaminophen (paracetamol) induced toxicity. The effect of *Eclipta alba* on biochemical parameters in control and acetaminophen induced hepatotoxic rats. Experimentally it has been shown to rise when acetaminophen (paracetamol) are given to rats. Administration of acetaminophen (750mg/kg; bodyweight), after 18 hours of intoxication resulted a significant ($P<0.05$) elevation of hepato-specific serum markers SGOT, SGPT, SALP, bilirubin and total protein in acetaminophen treated group, in comparison with the normal control group. On administration of CEEA and MEEA and Silymarin at the dose of 25mg/kg the level of these enzymes were found retrieving towards normalcy. While as at the 250mg/kg b.wt dose of *eclipta alba* methanol Extract showed highest percent of reduction in the cholesterol level (49.71%) followed by chloroform extract (27.16%) compared to acetaminophen treated rats. At 500mg/kg b.wt methanol extract showed (44.55%) and chloroform extract (23.91%) respectively. It should be noted that in all cases acetaminophen and plant extracts could not significantly changes the blood glucose level of rats

Keywords: Liver, *Eclipta alba* L, Acetaminophen, Biochemical parameter and Rattus *rattus*

1. Introduction
The liver is an organ of prime importance and plays a significant role not only in metabolism and detoxification of exogenous toxins and therapeutic agents, but also in the bio-regulation of blood coagulation and immuno-modulation. The liver is an important organ in the metabolic homeostasis of the body. However, due to its metabolic features and localisation, it is very vulnerable to toxic effects of xenobiotic, which can induce several steps of liver damages– from inflammatory to fibrotic processes. Impairment of the liver generally occurs from excessive exposure to xenobiotic, alcohol, chemotherapeutic agents, virus and protozoan infections. Depending upon the severity of the hepatic cell injury, viral acute hepatitis can lead to chronic hepatitis, which if left untreated can result in cirrhosis or malignant lesions. Antioxidants have also been proposed as therapeutic agents to counteract liver diseases, since reactive species are known to play a crucial role in liver diseases induction and progression. Additionally, because plant compounds are xenobiotics, they can induce toxicity to the liver, which highlight the importance of performing studies with liver cells. Moreover, possible enzyme and protein induction conferred by these products could provide an opportunity to mechanisms of interaction with other important drugs.
From the mechanisms behind liver cells injury, glutathione takes a vital place. The liver is the human organ with highest concentrations of glutathione. More than in other tissues, the levels of this intracellular antioxidant are very important in the protection against oxidative stress originated in the liver cells. Also, a drastic decrease of the reduced form of this tripeptide is known to be the central step in a cascade of events that culminate in extensive cell damage and death. Moreover, besides the detoxifying function of cellular glutathione, the recognition that this thiol can modulate signal transduction processes has recently been increasing. (Habig *et.al,* 1974) [17].
The liver is also very important in the regulation of the plasma levels of glucose. In the diabetes disease, which is characterised by a hyperglycaemic situation originated by a deregulation of insulin function and/or secretion, liver cells did not do their work properly.

Diabetes mellitus attained epidemic proportions in many countries and the available medication is far from resolving the problem, which makes it as a major public health concern. The liver is one of the potential targets for the treatment of diabetes and plants are currently again being used in the search for new possible drugs for its management. Diabetes is also associated with several complications where oxidative stress is known to be implicated, which make plant antioxidants interesting compounds to be researched against this chronic disease.

Chemical constituents of eclipta alba L.

Eclipta alba (L.) (syn. *Eclipta prostrata* L.), commonly known as False Daisy, and bhringraj, is a plant belonging to the family *Asteraceae*. Root well developed cylindrical, grayish. It is also named 'kehraj' in Assamese and karisalan kanni in Tamil. Floral heads 6-8 mm in diameter, solitary, white, achene compressed and narrowly winged. It grows commonly in moist places as a weed all over the world. It is widely distributed throughout India, China, Thailand, and Brazil (Bopanna *et.al* 1997) [6]. In *ayurvedic* medicine, the leaf extract is considered a powerful liver tonic, rejuvenative, and especially good for the hair. A black dye obtained from *Eclipta alba* is used for dyeing hair.

Eclipta alba also has traditional external uses, like athlete foot, eczema and dermatitis, on the scalp to address hair loss and the leaves have been used in the treatment of scorpion stings (Chandra *et.al* 1987) [9]. It is used as anti-venom against snakebite in China and Brazil. It is reported to improve hair growth and colour (Kritikar *et.al* 1975 and Chopra *et.al* 1955) [27, 11]. In Ayurveda the plant is considered a *rasayana* for longevity and rejuvenation. Recent studies have shown that it has a profound anti hepato-toxic activity. A cardio depressant activity was also observed in it when used for hepatic congestion (Bhattachary *et.al* 1997) [4]. A complete symptomatic relief in epigastric pain, nausea and vomiting in ulcer patients has also been observed (Puri, 2003) [42]. Also it is one among 10 flowers called as 'Dasapushpam' (Ten auspicious flowers) in Kerala, the southern state in India. The expressed leaf juice, applied along with honey, is a popular remedy for catarrh in infants. A preparation obtained from the leaf juice boiled with sesame or coconut oil is used for anointing the head to render the hair black and luxuriant (Sashi, 2000) [50]. An oil prepared with *amla, bhringraj* and sometimes with *brahmi* is well known in India as Amla Bhringraj oil, which is said to blacken the hair. Plant is rubbed on the gums in toothache and applied with a little oil for relieving headache and with sesame oil in elephantiasis. Roots of *Eclipt aalba* are emetic and purgative (Shastri *et. al* 1994) [55]. In Taiwan, entire plant is used as a remedy for the treatment of bleeding, haemoptysis, haematuria and itching, hepatitis, diphtheria and diarrhea (Gupta *et.al* 1999) [16]; in China, as a cooling and restorative herb, which supports the mind, nerves, liver and eyes. The leaf extract is considered to be powerful liver tonic, rejuvenative, and especially good for the hair (Murray *et.al* 2000) [36]. A black dye obtained from *Eclipta alba* is also for dyeing hair and tattooing. *Eclipta alba* also has traditional external uses, like athlete foot, eczema and dermatitis, on the scalp to address hair loss and the leaves have been used in the treatment of scorpion strings. It is used as anti-venom against snakebite in China and Brazil (Everitt *et. al* 2007) [13]. Tribal people of Rayalseema (Nagarajun *et al*., 1990) [37] (A.P., India) and others (Sahu *et al*., 1984) [46] apply extract of the bud and the herb respectively in sesame oil externally on forehead. Tribal people of Katra (J & K India) apply leaf juice with Neem oil for the growth of hair (Sahu *et al*., 1984) [46] while people (Japan) apply water extract of the herb externally for the purpose of itching (Tanaka *et al*., 1980) [57]. Chenchu tribe (AP., India) uses juice of plant with coconuts oil externally as hair tonic (Reddy *et al*., 1988) [43]. People in Tamilnadu (India) use fresh unripe fruit juice with other herbs in olive oil externally to prevent premature graying of hair (Kumar *et al*., 1987) [28]. Kani tribal people (Kerala, India) use leaf extract boiled with coconut oil to deepen black color of the hair and to promote their growth (John, 1984) [21]. Decoction (Bangladesh; Atahara *et al*., 1990) [3] and Taiwan (Linn *et al*., 1990) [31] is used orally. Hot water extract with other three herbs is used orally in Tirupati (AP., India), 4g thrice a day in divided doses for infective hepatitis in children (Vedavathyk *et al*., 1995) [61]. People in Bihar (India) use pills prepared from the root paste in insanity (orally), 4 to 5 pills twice a day for 7 days (Jain *et al*., 1994) [20]. In Arab countries leaf juice is used orally as hepatic tonic (Schmucker *et al*., 1969) [51]. In Tamilnadu people use fresh leaves, in Dandakaranya (Saxena *et al*., 1981) [50] (India) they use herb orally in case of Jaundice. They use root juice orally in liver complains in N. Gujarat (Shah *et al*., 1985) [53] (India). People in Cannanore (Kerala, India) use leaf with other herbs orally in case of jaundice (Ramachandran, 1987) [43]. Tribal people of Chittoor (AP., India) use dry powder of the herb orally, 4gm for a week to cure jaundice along with *P.amarus*to rejuvenate the liver (Reddy, 1988) [43]. In Rayalseema (AP., India) theyuse juice of the herb with butter milk and curd twice a day orally, but no salt in case of jaundice (Nagaraju *et al*. 1990) [37], Decoction (Ar-Arab countries, Schmucker *et al*., 1969) [51] and Lf-Nigeria (Akah *et al*., 1995) [2] are used orally as purgative and laxative respectively. (Prevention of miscarriage):10 to 15 ml juice of herb with cow's milk per day are taken orally from early days of pregnancy to prevent miscarriage and to have safe delivery (Bhattarai, 1994) [5].

Various biological activities are possessed by *E. alba,* such as memory disorders treatment, general tonic, edema, fevers and rheumatic joint pains treatment, digestion, hepatitis, enlarged spleen, antioxidant activity and skin disorders (Chopra *et.al*, 1956; Karnick and Kulkarni, 1990; Karthikumar *et. al*, 2007) [10, 22, 29]. Wedelolactone is active principle compound of this liver disorder treating drug (Wagner *et.al*, 1986) [64]. It also exhibits Trypsin inhibitory effect (Samiulla *et.al*, 2003; Syed *et.al*, 2003) [48, 55], suppresses LPS-induced caspase-11 expression in cultured cells by directly inhibiting the IKK complex (Kobori *et al*, 2004) [25], treatment of cirrhosis of the liver and infectious hepatitis (Murphy *et al*, 1979) [35]. The shoot extract of *E. alba* showed antimicrobial (Kosuge *et.al*, 1985; Wiart *et.al*, 2004) [26, 65], antifungal activity (Venkatesan and Ravi, 2004) [62] and weak cytotoxicity against the M-109 cell lines by alkaloids Verazine (Abdal Kadar *et.al*, 1998) [1], antiviral activity against Ranikhet disease virus (Khin *et.al*, 1978), effective against internal and external parasites (Lans *et.al*, 2001) [30] *G. intestinal is* anti bacterial (Kumar *et al.*, 2007) [29].

Since *E.alba,* are a weed /herb growing in dump, moist puddles distributed in the tropical and subtropical regions of the world. So besides ethno botanical evidence, it can be hypothesized that plants which survive in media rich in microbes most likely be possessing antimicrobial principles.

Hepato-toxicity and its adverse effects

Hepato-toxicity is a word derived from hepatic toxicity and refers to damage that is caused to the liver due to chemical driven damage. Chemicals that cause damage to the liver are referred to as hepatotoxins and include carbon tetrachloride, alcohol, dantrolene sodium, valproic acid, and isonicotinic acid hydrazide. This sort of damage can be a result of side effects due to certain types of medicines but may also be a result of certain natural chemicals and chemicals employed in industry and laboratories. The most common form of liver poisoning observed in western countries due to medication is from that caused by paracetamol poisoning known as acetaminophen. Sometimes certain medicines are not poisonous or toxic in their compounds but do become toxic when broken down by the liver. Liver plays a vital role in the metabolism and elimination of various exogenous and endogenous compounds. As a result of its continuous involvement, it is susceptible to toxic injuries caused by certain agents and any damage to hepatic cells disturbs body metabolism. In recent times lots of interest has been generated to find out a natural remedy for hepatic disorders caused by toxins like alcohol and hepatitis virus (Patel *et.al,* 1998) [40]. The agent should protect against such damage, especially of one which facilitates regeneration by proliferation of parenchymal cells after damage and arrest growth of fibrous tissue. There is no remedy for liver diseases which are so prevalent in the population. The treatment is mainly symptomatic. (Rege *et. al,* 1984) [44] Scientists and some industrialists deliberated on various prospective plant remedies for ailments of liver disorder management. In the decade 70s, the world scientific community concentrated on a herbal plant Vincarosea. Then in 80s the attention was focused on Panax ginseng. Now, the news of multifarious activities of the Neem tree indicates that it may become centre for research in 90s. Indian Council of Medical Research, New Delhi, in its revived research on traditional medicine, had adopted liver diseases as one among six thrust areas and for multidisciplinary study. Screening of active constituents from Kutki (Picrorhiza-kurroa), Bhoomy-amalaki (Phyllan-thusniruri) have shown marked protection against jaundice. Hepatitis continues to be a major health problem in urban areas in India, and several studies in viral hepatitis were under investigation by the ICMR. For example, extracts of milk thistle (Silybum-marianum) fruits under investigation for the treatment of alcoholic hepatitis. According to Indian Society of Gastroenterology, Mulethi (Glycyrrhizaglabra) prevents multiplication of viruses inside liver cells. The disorder of liver may be acute or chronic hepatitis (inflammatory liver diseases), hepatosis (non-inflammatory liver diseases) and liver cirrhosis (fibrosis of the liver). Liver enzymes act as an index of sub-clinical hepatic damage. Serum glutamic pyruvic transaminase (SGPT), serum glutamic oxaloacetic pyruvic transaminase (SGOT), Serum lactic dehydrogenase (LDH) and Serum alkaline phosphatase are reported as an index of hepatic injury and cholestasis (Doreswamy *et.al,* 1995) [12].

In the present study, we have been planning to elucidate hepato-protective potential of *Eclipta alba* against chemically induced hepato-carcinogenesis in *Rattus rattus*.

Material and Methods

Collection and Identification of Plant material

The plants of *eclipta alba L.* were collected in April directly from the Nehru Nagar surroundings of Bhopal Madhya Pradesh, India and authenticated with the help of botanist at college and voucher samples were preserved for reference in the herbarium. The collected leaves were dehydrated in hot air dryer at 40 ± 5ºC (one batch) and were stored for further experiments. The plant part used for study are given in (Table 1).

Table 1: Showing Plant material and its part used.

Plant Name	Common name	Family	Parts used	Month of collection	Season of collection
Eclipta Alba	False Daisy or Bhringraj	Asteraceae	Leaves	April-May.	Summer

Fig 1: *Eclipta alba.* L.

Botanical Classification of *Eclipta alba* Lin.

Kingdom	:	Plantae
Phylum	:	Angiosperms
Subphylum	:	Eudicots
Order	:	Asterales
Family	:	Asteraceae
Genus	:	*Eclipta*
Species	:	*alba*

Geographical distribution (Caton *et al,* 2004) [8]

Eclipta alba L. is found in both tropical and sub-tropical habitats in Asia: China (including Taiwan), Japan, and Korea. South and Southeast Asia: Bangladesh, India, Indonesia, Cambodia, Lao PDR, Malaysia, Nepal, Pakistan, Philippines, Sri Lanka, Thailand, and Vietnam. Rest of the world: Angola, Arabian Peninsula, Argentina, Australia, Brazil, Colombia, Costa Rica, Cote d'Ivoire, Cuba, Egypt, Fiji, Ghana, Iraq, Mexico, Peru, Portugal, Puerto Rico, Rhodesia, Sudan, Surinam, Trinidad, United States of America (including Hawaii), Zambia, and Zimbabwe.

Morphology (Caton *et al,* 2004) [8]

A prostrate or reclining to erect, often branched, annual or perennial herb, 30-100cm tall. Stem is cylindrical, green or purplish, rooting at basal nodes, and often covered with long white hairs. Leaf is oblong to lance-shaped, opposite, sessile or short-stalked, with more or less coarse hairs; margins entire

or slightly toothed, up to 2-16 cm long. In florescence terminal and axillary, about 1cm across, white or cream, on peduncles to 7cm long. Fruit is achene, densely warted, either brown or black, 2-3mm long.

Biology and Ecology: (Caton *et al*, 2004) [8]
Wide spread and adapted to a range of environments. Found in poorly drained wet areas, saline conditions, along streams, in drains and canals of irrigated lowland rice paddies, in waste areas, and in upland fields. A single plant can produce as many as 17,000 seeds; germination affected by light, moisture level, pH, and temperature, but seeds have no dormancy.

Preparation of Extract (Harbone, 1988)
The whole plant was dried under shade and then powered with a mechanical grinder to obtain a coarse powder, which was then subjected to successive extraction in a maceration apparatus using petroleum ether (60-80^{0}C), chloroform and methanol. Solvent elimination under reduced pressure afforded the chloroform extract (2 % v/w yield) and methanol extract (17 % v/w yield) respectively. The resulting chloroform and methanol extracts were then used for hepato protective and *in vivo* antioxidant studies.

Experimental animals
Wistar albino rats (150-200g) and Swiss albino mice (20-25g), of either sex roughly the same age (8-10 weeks), obtained from the Experimental Animal Care Centre, Division of Pharmacology, IIIM(CSIR), Jammu were used. The animals were housed under constant temperature (22 ± 2°C), humidity (55%) and light/dark conditions (12/12 h). They were provided with Purina chow and free access to drinking water *ad libitum.*

Experimental design: (Hiroshini *et.al*, 1987) [19]
Animals were randomized and divided into seven groups (I-VII) of six animals in each group. Group I served as untreated control and fed orally with normal saline 5ml/kg body weight daily for seven days. Group II rats were similarly treated as group I. Group III and IV were treated with 250mg and 500mg/kg body weight of the chloroform extract and Group V and VI were treated with the methanolic extract orally daily for seven days respectively. Animals of Group VII were fed with standard drug Silymarin 25mg/kg12; p.o daily for seven days. On the seventh day, Acetaminophen suspension was given by oral route; in a dose of 750mg/kg body weight to all rats except the rats in group I. The biochemical parameters were estimated after an 18h fast following the last dose.

Biochemical analysis
The blood was obtained from all animals by puncturing retro-orbital plexus. The blood samples were allowed to clot for 45 min at room temperature. Serum was separated by centrifugation at 2500rpm at 30^{0}C for 15 min and utilized for the estimation of various biochemical parameters namely SGOT, SGPT (Rietman *et.al*, 1957) [45], SALP serum (Reitman *et.al*, 1957: Nobert, 1970, Godkar *et.al*, 2006) [45, 38, 14] bilirubin and total protein (Malloy and Evelyn, 1937) [33]. After collection of blood samples the rats indifferent groups were sacrificed and their livers were excised immediately and washed in ice cold normal saline, followed by 0.15 M Tris-Hcl (pH 7.4) blotted dry and weighed. A 10% w/v of homogenate was prepared in 0.15 M Tris-Hcl buffer. A part of homogenate after precipitating proteins with Trichloroacetic acid (TCA) was used for estimation of glutathione. The rest of the homogenate was centrifuged at 1500 rpm for 15 min at 40^{0}C.

Estimation of SGPT (Reitman *et.al*, 1957: Nobert, 1970, Godkar *et. al*, 2006) [45, 38, 14]
Intended Use
This reagent kit is intended for *in-vitro* quantitative determination of SGPT (ALT) activity in serum/plasma.

Estimation of SGOT
Estimation of serum bilirubin, total protein
Serum total bilirubin level was estimated based on the method of (Malloy and Evelyn, 1937) [33]. Serum total protein level was estimated based on the method of Gornall *et.al*, 1949 [15].

Determination of serum Glucose, Phosphorus, Lipid, A/B ratio and Cholesterol
Tissue parameters like Glucose, phosphorus, lipid and A/G ratio and Cholesterol were determined by using Standard kits supplied by Span Diagnostics Ltd., Surat, India.

Results
The Present study was focused at evaluating the hepatoprotective effect of chloroform and methanol extract of *Eclipta alba L.* by acetaminophen (paracetamol) induced liver damage in rats. The chloroform and methanol extracts were studied for their hepato-protective effects on acetaminophen (750mg/kg) induced acute liver damage on Wister albino rats. The degree of protection was measured by using biochemical parameters.

Extraction
The *Eclipta alba,* leaves powder was subjected to maceration. The percentage yield, colour, consistency and solubility in water were noted (Table 2)

Table 2: The percentage yield, color, consistency and solubility in water of *Eclipta alba* Extracts.

Plant part used	Extract	%yield	Color	Consistency	Solubility in water
Eclipta alba	Methnol	17%	Green	Powder	Freely soluble
Leaves	Chloroform	2%	Green	Semi-solid	Freely Soluble

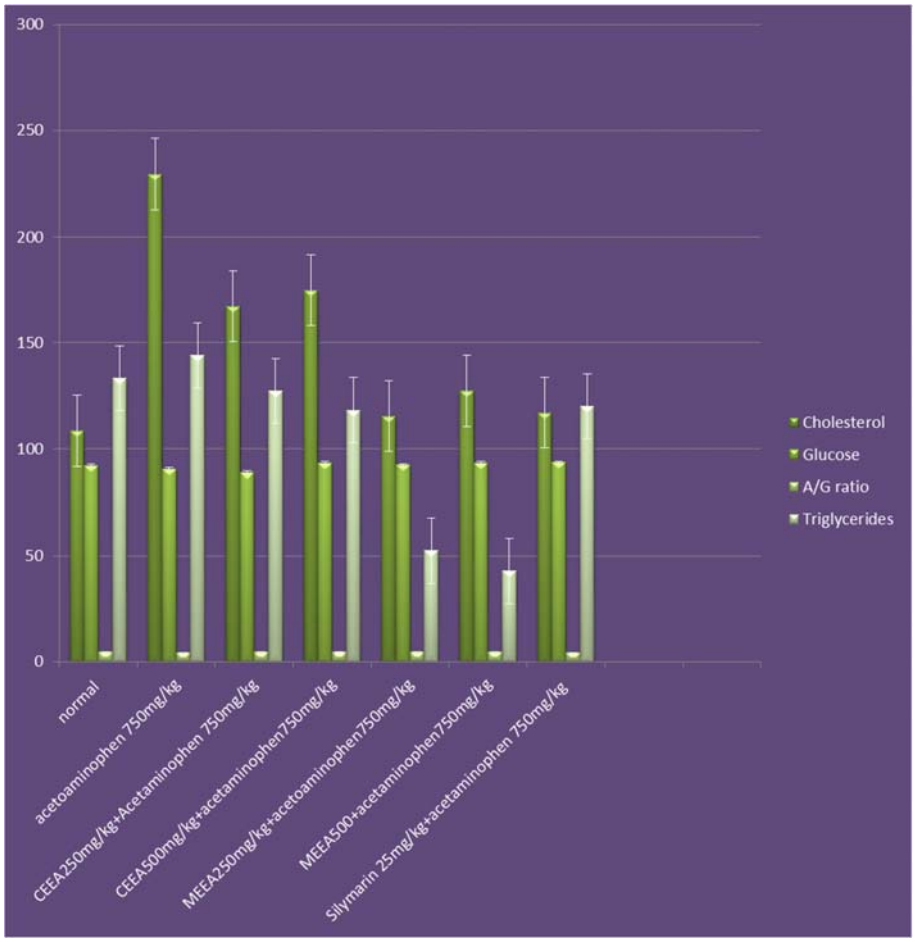
Determination of Hepato-protective activity of *Eclipta alba* L. Leaves extract
Biochemical Parameters
The serum glutamic oxallo transaminase (SGOT) and serum pyruvic oxallo transaminase (SGPT) are two enzymes, which are normally present inside the cells. When the cell is damaged these enzymes leak out and when a large number of cells are damaged their level in the blood will rise. It has been shown to rise clinically in myocardial infarction and in cases of liver damage. Experimentally it has been shown to rise when acetaminophen (paracetamol) are given to rats. Acetaminophen causes damage to liver cells and necrosis of liver, in adequate doses. The effects of CEEA and MEEA on serum transaminase, alkaline phosphatase, bilirubin and total

protein levels in Acetaminophen (paracetamol) induced liver damage in rats are summarized in (Table 3). Administration of acetaminophen (750mg/kg; bodyweight), after 18 hours of intoxication resulted a significant (P<0.05) elevation of hepato-specific serum markers SGOT, SGPT, SALP, bilirubin and total protein in acetaminophen treated group, in comparison with the normal control group. On administration of CEEA and MEEA (Group III to VI, Table-3) and Silymarin at the dose of 25mg/kg (Group VII, Table-3) the level of these enzymes were found retrieving towards normalcy. Cholesterol has been attributed to the damaged structural integrity of the liver, because they are cytoplasmic in location and released into circulation after cellular damage. At the 250mg/kg b.wt dose of *eclipta alba* methanol Extract showed highest percent of reduction in the cholesterol level (49.71%) followed by chloroform extract (27.16%) compared to acetaminophen treated rats. At 500mg/kg b.wt methanol extract showed (44.55%) and chloroform extract (23.91%) respectively. It should be noted that in all cases acetaminophen and plant extracts could not significantly changes the blood glucose level of rats (table 3) However, there was no significant effect on the serum A/G ratio. Liver injury causes the accumulation of abnormal amounts of fats, predominantly triglycerides in the parenchymal cells. Triglycerides accumulation can be thought of as resulting from an imbalance between the rate of synthesis and the rate of release of triglycerides by the parenchymal cells into the systemic circulation. The elevated plasma triglycerides levels observed might have been partially due to lipoprotein lipase. Modest hypertriglyceridemia occurs in association with alcohol, virus and drug induced hepatitis. In the Present study acetaminophen significantly increased the serum triglyceride levels indicating liver injury while treatment with methanol extract as well as chloroform extract of *E.alba* in both doses significantly decreased triglyceride levels, their lowering effect was more potent than silymarin. Thus concluding hepato-protective activity of *Eclipta alba* against hepato-toxicity of acetaminophen.

Table 3: Effect of Chloroform (CEEA) and Methanol (MEEA) extract on biochemical parameters.

Groups	Cholesterol (mg/dl)	Glucose (mg/ dl)	A/G Ratio (g/dl)	Triglyceride (mg/dl)
Group I Normal	108.66 ± 1.11	92.33 ± 3.48	4.6±0.3	133.3±19.2
Group II Acetaminophen 750mg/kg	229.67 ± 0.75	90.66 ± 2.45	4.36±0.4	144±14.4
Group III CEEA 250mg + Acetaminophen	167.29± 0.73	89.08 ± 1.20	4.6±0.3	127.3±17.7
Group IV CEEA 500mg + Acetaminophen	174.75±1.17	93.5±2.60	4.50±0.7	118.2±16.3
Group V MEEA 250mg+ Acetaminophen	115.50±5.79	92.50±2.09	4.6±0.6	52.46±8.9
Group VI MEEA 500mg+Acetaminophen	127.33±0.71	93.50±2.99	4.6±0.5	42.88±8.7
Group VII Silymarin25mg+ Acetaminophen	117.1±8.71	93.55±2.60	4.35±0.4	120.1±8.4

All values are Mean ± SEM, P<0.05 was considered significant with respect to hepatotoxic control



Effect of *Eclipta alba L.* extracts on glucose cholesterol A\G ratio and triglycerides

Discussion

In the present study, we have been planning to elucidate hepato-protective potential of *Eclipta alba* against chemically induced hepato-carcenogenasis in *Ratus ratus*. Treatment of diseases associated with the liver is very vital, and must be done with importance and extensive care. Many herbal remedies for liver diseases are known but only a few of them have been pharmacologically assessed for their efficacy. It is very important to assess natural products for their efficacy in the treatments they are used for. It is especially very important to assess remedies for liver diseases due to the liver's fragility and relation to other vital organs, and yet it's numerous vital roles detrimental to the survival of a person.

In recent times, due to economic factors, people are in need of available, easily accessible and less costly medication, even with the slightest knowledge of efficacy, and minimum idea of toxicity. It is believed by most people that since herbal remedies are natural, they are non-toxic. Toxicity of natural remedies have however been reported. Even scientifically proven hepato protective plant was found to contain hepatotoxins as well (Bramanti *et.al*, 1978; MacGregor *et.al*, 1989; Oshima, 1995) [7, 32, 39]. Thus, work on hepato protective herbal remedies remain a challenge (Schuppan *et.al*, 1999) [52].

Paracetamol (acetaminophen) is a commonly and widely used analgesic and antipyretic agent. Hepato toxic doses of acetaminophen deplete the normal levels of hepatic glutathione, when NAPQI covalently binds to cysteine groups on proteins to form 3-(cystein-S-yl) acetaminophen adducts (Timenstein and Nelson, 1989) [59]. The glutathione protects hepatocytes by combining with the reactive metabolite of paracetamol thus preventing their covalent binding to liver proteins (Vermsulen *et.al,* 1992) [63].

In living systems, liver is considered to be highly sensitive to toxic agents. The study of different enzyme activities such as SGOT, SGPT, SALP, total bilirubin and total protein have been found to be of great value in the assessment of clinical and experimental liver damage (Vaishwanar and Kowale, 1976) [60]. In the present investigation it was observed that the animals treated with acetaminophen resulted in significant hepatic damage as shown by the elevated levels of serum markers. These changes in the marker levels will reflect inhepatic structural integrity. The rise in the SGOT is usually accompanied by an elevation in the levels of SGPT, which play a vital role in the conversion of amino acids to keto acids (Sallie *et.al*, 1999) [47]. The pre-treatment with CEEA and MEEA, both at the dose of 250mg/kg and 500mg/kg, significantly attenuated the elevated levels of the serum markers. The normalization of serum markers by CEEA and MEEA suggests that they are able to condition the hepatocytes so as to protect the membrane integrity against acetaminophen induced leakage of marker enzymes into the circulation. The above changes can be considered as an expression of the functional improvement of hepatocytes, which may be caused by an accelerated regeneration of parenchyma cells. Serum ALP and bilirubin levels, on the other hand are related to hepatic cell damage. Increase in serum level of ALP is due to increased synthesis in presence of increasing billiary pressure (Moss and Butterworth, 1974) [34]. Effective control of bilirubin level and alkaline phosphatase activity points towards an early improvement in the secretory mechanism of the hepatic cell.

In the present study the elevation of the blood glucose and A/G ratio levels were not significant by the treatment of *Eclipta alba* extracts. The rise in the serum levels of AST, ALT, Triglycerides and cholesterol has been attributed to the damaged structural integrity of the liver, because they are cytoplasmic in location and released into circulation after cellular damage (Moss and Butterworth, 1974) [34]. When rats were treated with acetaminophen it induces hepatotoxicity by metabolic activation, therefore it selectively causes toxicity in the liver cells maintaining semi-normal metabolic function. Acetaminophen is metabolically active by the cytochrome P450 dependent mixed oxidases in the endoplasmic reticulum and induced lipid peroxidation. These result in changes of structure of endoplasmic reticulum and other membrane, loss of metabolic enzyme activation, reduction of protein synthesis and loss of glucose-6-phosphate activation and leading to liver injury. The treatment of *Eclipta alba* methanol extract showed potential antiheptotoxic effect. These results were in accordance with (Tabassum and Agarwal, 2004; Trirumalai *et.al*, 2011) [56, 58].

At the 250mg/kg b.wt dose of *eclipta alba* methanol Extract showed highest percent of reduction in the cholesterol level (49.71%) followed by chloroform extract (27.16%) compared to acetaminophen treated rats. At 500mg/kg b.wt methanol extract showed (44.55%) and chloroform extract (23.91%) respectively. It should be noted that in all cases acetaminophen and plant extracts could not significantly changes the blood glucose level of rats.

Acetaminophen significantly increased the serum triglycerides levels. Methanol extract as well as chloroform extract of *eclipta alba* in both doses significantly decreased triglycerides levels, their lowering effect was more potent than silymarin. However, there was no significant effect on serum A/G ratio. The present concluded that *Eclipta alba* has profound and significant protection against Acetaminophen (paracetamol)-induced toxicity.

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