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IN VIVO HEPATOPROTECTIVE EFFECT OF ETHANOLIC EXTRACT OF CAPSICUM ANNUUM L. RED VARIETY AGAINST PARACETAMOL INDUCED HEPATOTOXICITY IN MALE WISTAR ALBINO RATS

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ABSTRACT
This study was designed to evaluate the hepatoprotective activity of ethanolic extract of *Capsicum annuum* L. Red variety (*C. annuum*) against paracetamol induced liver damage in male wistar albino rats. The ethanolioc extract was administered orally to the wistar albino rats with hepatotoxicity induced by paracetamol. Silymarin was given as reference standard. The plant extract was effective in protecting the liver against the injury induced by paracetamol in rats. The liver injury was induced by paracetamol@750mg/kg b.w orally. The rats were divided in five groups. Rat of groupG1 served healthy control receive no treatment. G2 served as toxic control and receive paracetamol orally @750mg/kg at every 72h interval for 21d. G3 received with standard silymarin 50mg/kg for 21d and simultaneously administered paracetamol. G4 received with ethanolic extract of *C. annuum* 250mg/kg for 21d and simultaneously administered paracetamol. G5 received ethanolic extract of *C. annuum* 500mg/kg for 21d and simultaneously administered paracetamol. Various biochemical parameters like lipid content, free fatty acids, triglycerides, phospholipids, glycoprotein, hexose, hexosamine, sialicacid and glycogen were estimated liver tissue of treated and untreated animals. In serum, total protein, albumin, aspartate transaminase, alanine transaminase, alkaline phosphatase and total bilirubin were analyzed using commercially available test diagnostic kits. This was evident from significant reduction in serum enzymes alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP). Our results confirmed that the ethanolic extract of *C. annuum* possesses hepatoprotective activity against paracetamol induce hepatotoxicity rats.

KEYWORDS: Hepatoprotection, Paracetamol, *Capsicum annuum* L, Silymarin.

1.0 INTRODUCTION
Liver is one of the vital organs in human body and principal site for enhanced metabolism and excretion. It is responsible for the detoxification of various drugs and xenobiotics in the body. Liver diseases have become one of the major causes of morbidity and mortality all over world. From among, Drug Induced Liver Injury (DILI) is one of the most common causative factor that poses a major clinical and regulatory challenge (Russmann *et al.*, 2009). Paracetamol (PCM) also known as Acetaminophen, taken in overdose can cause severe hepatotoxicity and nephrotoxicity (Vermeulen *et al.*, 1992). Drug induced liver toxicity is major health problem that challenges not only healthcare professionals but also the pharmaceutical industry and also drug regulatory agencies (Ajith *et al*., 2002). Due to enormous side effects of allopathic medicine, today the world is returning toward the natural options for diseases treatment. About 80% of the world population relies on traditional medicine which is predominantly based on the plant materials (WHO, Manila 1993). Currently, 25% of all modern medicines are directly or indirectly derived from higher plants. Silymarin is a standardized extract of the milk thistle (*Silybummarianum*) have been used for more than 2000 years to treat liver and gall bladder disorders, including hepatitis, cirrhosis and jaundice and to protect the liver against poisoning from chemicals, environmental toxins, snake bites, insect stings, mushroom poisoning and alcohol (Kren and Walterova, 2005). *C. annuum* is commonly known as Chilli pepper and it is most widely cultivated throughout the world and belongs to the family Solanaceae. The fruit is proved to possess antiulcer activity, anticancer activity, antioxidant activity, hepatoprotective and radical scavenging properties (Ahmed *et al.*, 2012). The methanolic seed extract of the plant reported antiobesity activity in 3T3-

L1 adipocyte (Lakshmi *et al.*, 2013). Hence, an attempt was made to evaluate the antioxidant and hepatoprotective potential of *C. annuum* fruits against paracetamol induced liver damage in male wistar albino rats.

2.0 MATERIALS AND METHOD

2.1 Drugs and chemicals

PCM and Silymarin were gifted from Microlabs, Bengaluru, India. Kits for the estimation of selected biochemical parameters were of Erba Diagnostics, Germany. All other chemicals used in this study were of analytical grade.

2.2 Sample collection and Extraction

Fresh fruits of organically farming *C. annuum* red variety were purchased from a Srivatsa Organic Farms, Coimbatore, Tamil Nadu.

2.3 Quantification of Pigments

2.3.1 Extraction and estimation of anthocyanin of *C. annuum*

20 mg of fresh red variety of *C. annuum* was extracted using mortar and pistle with 2.5 ml of an 80% ethanol and the supernatant was collected by centrifugation and stored at 4ºC for further studies. Total anthocyanin content was determined by the modified method given by Giusti and Wrolstad (2001).

2.3.2 Extraction and estimation of lycopene

The lycopene from *C. annuum* pulp was extracted using acetone and quantification was done according to the method of Sadasivam and Manickam, (2009).

2.4 Evaluation of hepatoprotective effect of ethanolic extract of *C. annuum*

2.4.1 Processing of red variety of *C. annuum*

The fresh fruits of *C. annuum* were washed in running tap water and wiped off to avoid dryness. The fruits were finely chopped into pieces and dried under shade. The shade dried *C. annuum* pieces were powdered using a mechanical grinder and used for extraction.

2.4.2. Ethanolic extraction of *C. annuum* red variety

The air dried, powdered *C. annuum* was extracted in Soxhlet extractor with ethanol (25g in 450-500 ml) for 6-8 hrs. The ethanolic *C. annuum* solution obtained was concentrated in an evaporator and was dried to remove even the final traces of ethanol. The percentage of yield was calculated. The hepatoprotective potential was evaluated by dissolving it in distilled water right before use.

2.5 Experimental animals

Male albino wistar rats (150-200 g) of the study were procured from the small animals breeding station, Mannuthy, Kerala, India. They were housed in polypropylene cages (38 x 23 x 10cm) with not more than six animals per cage and maintained under standard environmental conditions (14h dark /10h light cycles; temp 25±2°C; 35-60% humidity, air ventilation) and were fed with standard pellet diet (M/s. Hindustan Lever Ltd., Mumbai, India) and fresh water *ad libitum*. The animals were acclimatized to the environment for two weeks prior to experiment use. Animals were fasted over night before the experimental schedule, but have free access for water *ad libitum.*

2.6 Experimental design

The hepatoprotective was investigated against paracetamol induced hepatotoxicity in rat model following the method of Araya *et al*., (1987). The rats were segregated into 5 groups of six animals each. The experiment was designed as follows.

Group I: Control animals received the vehicle viz. normal saline (2 ml/kg).

Group II: Received paracetamol (750 mg/kg) at every 72 h interval for 21 days.

Group III: Received silymarin (50 mg/kg) for 21 days and simultaneously administered paracetamol 750 mg/kg every 72 h.

Group IV: Received ethanolic extract of *C. annuum* (250 mg/kg) for 21 days and simultaneously administered paracetamol 750 mg/kg every 72 h.

Group V: Received ethanolic extract of *C. annuum* (500 mg/kg) for 21 days and simultaneously administered paracetamol 750 mg/kg every 72 h.

24hrs after the dose of paracetamol administration, blood samples were collected from all groups by puncturing retro-orbital plexus. The blood samples were allowed to clot for 45 min and the serum was separated by centrifugation at 2500 rpm for 15 min and analyzed for various biochemical parameters. After collection of blood samples, the rats were sacrificed and their liver excised, rinsed in ice cold normal saline followed by 0.15 M TrisHCl (pH 7.4), blotted dry and weighed.

2.7 Biochemical Analysis

Various biochemical parameter like lipid content(Folch *et al*., 1957), free fatty acids (Hron and Mehana., 1981), triglycerides (Rice., 1970), phospholipids (Rouser *et al.*, 1970), glycoproteins (Pari and Karthikesan., 2009), hexose (Niebes., 1972), hexosamine (Wagner., 1979), sialicacid (Niebes., 1972) and Glycogen
(Maiti *et al*., 2004) were estimated liver tissue of treated and untreated animals.

2.8 Estimation of serum liver markers

In serum, total protein, albumin, aspartate transaminase, alanine transaminase, alkaline phosphatase and total Bilirubin were analyzed using commercially available test diagnostic kits.

2.9 Statistical Analysis

The results were expressed as mean ± SD. Statistical significance of differences between groups was determined by one way ANOVA followed by LSD test. *P* values of <0.05 is considered significantly different. The calculations were performed using SPSS version 17.

3.0 RESULTS

3.1 Quantification of pigments in *C. annuum* red variety

The lycopene and anthocyanin content was determined in *C. annuum* red variety according to the standard procedure. The lycopene content was found to be 4.69 ± 0.01 mg/g tissue and the anthocyanin was found to of 1.52 ± 0.09 mg/100 g dry matter.

3.2 Effect of ethanolic extract of *C. annuum* red variety on liver weight

Paracetamol induction resulted in an increase in the weight (3.56 ± 0.28 g/100g b.w.) of the liver tissue compared to the control group (2.06 ± 0.19 g/100g b.w.) animals. A significant (P < 0.001) increase in the liver weight of induced group animals were observed compared to the un-induced, untreated control animals. The standard drug, silymarin had the efficacy to revert back the liver weight to normal levels (2.39 ± 0.31 g/100g b.w.) in a significant manner (P< 0.001). In a similar way, the ethanolic extract of *C. annuum* also lead to a decrease in the liver weight in a dose dependent manner (200 mg/kg b.w. - 2.99 ± 0.15 g/100g b.w.; 400 mg/kg b.w. - 2.29 ± 0.28 g/100g b.w.) (Table 1).

3.3 Effect of ethanolic extract of *C. annuum* red variety on glycogen in liver tissue

In concern with glycogen storage level in the liver tissue,a significant (P< 0.001) decrease in the ability of the liver to store glycogen was highly decreased in paracetamol induced group (24.73 ± 1.07 mg/g tissue) of animals compared to the control group animals (49.18 ± 2.15 mg/g tissue). The standard drug, silymarin had the potency to significantly increase (P< 0.001) the ability of liver to store glycogen (48.03 ± 1.17 mg/g tissue). Among the low and high dose of the ethanolic extract of *C.annuum*, the high dose was equally potent to that of the standard drug which showed the glycogen storage to be 48.10 ± 1.29 mg/g tissue (Table 1).

Table 1: Liver weight and glycogen concentration in liver tissue of control and experimental groups.

Groups	Liver weight g/100 g b.w	Glycogen mg/g tissue
Control	2.06 ± 0.19	49.18 ± 2.15
Induced	3.56 ± 0.28###	24.73 ± 1.07###
Standard (Silymarin - 50 mg/kg b.w.)	2.39 ± 0.31***	48.03 ± 1.17***
Low dose (250 mg/kg b.w.)	2.99 ± 0.15***	38.78 ± 0.87***
High dose (500 mg/kg b.w.)	2.29 ± 0.28***	48.10 ± 1.29***

#Change in activities at P<0.05 when induced compared to control, ## P<0.01, ### P<0.001; *Change in activities at P<0.05 when standard, low dose and high dose compared to induced, ** P<0.01, *** P<0.001; Values are expressed as mean ± SD (n=6).

3.4 Effect of ethanolic extract of *C. annuum* red variety on lipid content of liver tissue

The lipids determined in the liver tissue of all the group of animals were free fatty acids, phospholipids and triglycerides. Paracetamol had the ability to significantly increase (P< 0.001) the lipid content in the liver tissue when compared to the control group. The free fatty acids, phospholipids and triglycerides contents in the control group animals were found to be 1.51 ± 0.02 mg/g tissue, 0.09 ± 0.01 mg/g tissue and 2.79 ± 0.03 mg/g tissue whereas their levels were elevated to 4.95 ± 0.03 mg/g tissue, 1.41 ± 0.07 mg/g tissue and 7.33 ± 0.13 mg/g tissue in the paracetamol induced groups.

The standard drug, silymarin significantly (P< 0.001) protected the liver tissue by decreasing the lipid levels to normal. It brought back the lipid levels almost closer to the ones in the control group animals. The free fatty acids, phospholipids and triglycerides were found to be 1.56 ± 0.08 mg/g tissue, 0.10 ± 0.02 mg/g tissue and 2.81 ± 0.24 mg/g tissue respectively. On treatment with the ethanolic extract of *C. annuum*, there was a significant decrease (P< 0.001) in the lipid levels of liver tissue in a dose dependent manner. Both the low dose and high dose of the extract established protection against increased lipid levels which was observed due to paracetamol induction. The high dose of ethanolic extract of *C. annuum* reduced the free fatty acids, phospholipidsand triglycerides contents to 1.76 ± 0.05 mg/g tissue, 0.14 ± 0.02 mg/g tissue and 3.44 ± 0.40 mg/g tissue correspondingly (Table 2).

Table 2: The concentration of free fatty acids, phospholipids and triglycerides in liver tissue of control and experimental groups.

Groups	FFA mg/g tissue	PL mg/g tissue	TG mg/g tissue
Control	1.51 ± 0.02	0.09 ± 0.01	2.79 ± 0.03
Induced	4.95 ± 0.03###	1.41 ± 0.07###	7.33 ± 0.13###
Standard(Silymarin - 50 mg/kg b.w.)	1.56 ± 0.08***	0.10 ± 0.02***	2.81 ± 0.24***
Low dose (250 mg/kg b.w.)	2.24 ± 0.07***	0.67 ± 0.09***	5.76 ± 0.18***
High dose (500 mg/kg b.w.)	1.76 ± 0.05***	0.14 ± 0.02***	3.44 ± 0.40***

Change in activities at P<0.05 when induced compared to control, ## P<0.01, ### P<0.001; *Change in activities at P<0.05 when standard, low dose and high dose compared to induced, ** P<0.01, *** P<0.001; Values are expressed as mean ± SD (n=6). [FFA- Free Fatty Acids, PL – Phospholipids, TG- Triglycerides].

3.5 Effect of ethanolic extract of *C. annuum* red variety on glycoprotein content of liver tissue

The glycoproteins that were assessed include hexose, hexosamine and sialic acid in the liver tissue of all the group of animals are given in Table 3. A significant reduction ($P< 0.001$) in the level of glycoproteins were observed in the untreated paracetamol induced group compared to the normal control animals. The levels of hexose, hexosamine and sialic acid in the control group animals were found to be 866.36 ± 17.09 mg/g defatted tissue, 57.24 ± 1.08 mg/g defatted tissue and 6.36 ± 0.67 mg/g defatted tissue respectively. Their levels were reduced in the paracetamol induced animals to 589.55 ± 17.85 mg/g defatted tissue, 26.79 ± 1.36 mg/g defatted tissue and 3.38 ± 0.54 mg/g defatted tissue. Silymarin, the standard drug at a dose of 50 mg/kg b.w. significantly increased the glycoprotein levels to normal values. Their levels on treatment for a period of 21 days were increased to 821.24 ± 12.00 mg/g defatted tissue, 58.37 ± 1.35 mg/g defatted tissue and 6.66 ± 1.03mg/g defatted tissue. On treatment with ethanolic extract of *C. annuum* at a dose of 200 mg/kg b.w., the glycoprotein levels of the animals were raised to 660.91 ± 9.94 mg/g defatted tissue, 46.78 ± 1.75 mg/g defatted tissue and 4.44 ± 0.42mg/g defatted tissue. The high dose, 400 mg/kg b.w., significantly rose ($P< 0.001$) the levels of hexose, hexosamine and sailic acid to 720.15 ± 14.33 mg/g defatted tissue, 55.64 ± 2.86 mg/g defatted tissue and 5.86 ± 0.78mg/g defatted tissue respectively.

Table 3: The concentration of glycoproteins in liver tissue of control and experimental groups.

Groups	Hexose mg/g defatted tissue	Hexosamine mg/g deffated tissue	Sialic acid mg/g deffated tissue
Control	866.36 ± 17.09	57.24 ± 1.08	6.36 ± 0.67
Induced	589.55 ± 17.85###	26.79 ± 1.36###	3.38 ± 0.54###
Standard (Silymarin - 50 mg/kg b.w.)	821.24 ± 12.00***	58.37 ± 1.35***	6.66 ± 1.03***
Low dose (250 mg/kg b.w.)	660.91 ± 9.94***	46.78 ± 1.75***	4.44 ± 0.42*
High dose (500 mg/kg b.w.)	720.15 ± 14.33***	55.64 ± 2.86***	5.86 ± 0.78***

#Change in activities at $P<0.05$ when induced compared to control, ## $P<0.01$, ### $P<0.001$; *Change in activities at $P<0.05$ when standard, low dose and high dose compared to induced, ** $P<0.01$, *** $P<0.001$; Values are expressed as mean ± SD (n=6).

3.6 Effect of ethanolic extract of *C. annuum* red variety on protein, albumin and total bilirubin of serum

Paracetamol induction at a dose of 750 mg/kg b.w., for 21 days with time interval of 72 hrs significantly decreased ($P< 0.001$) the values of protein and albumin to 4.67 ± 0.42 mg/dl and 2.74 ± 0.14 mg/dl. Their levels in control group animals were 7.88 ± 0.52 mg/dl and 4.20 ± 0.07 mg/dl. Treatment with 50 mg/kg b.w. of standard drug, silymarin increased the protein and albumin levels. The ethanolic extract of *C. annuum* also had the potential to alter the protein and albumin value to near normalcy. Both the low and high dose significantly ($P< 0.001$) increased the protein and albumin values. The low dose was also equally potent as that of the high dose. The protein and albumin levels of low and high dose groups were found to be 7.32 ± 0.60 mg/dl and 4.44 ± 0.18 mg/dl; 7.92 ± 0.35 mg/dl and 4.47 ± 0.16 mg/dl respectively. Bilirubin serves as an important parameter in assessing the functions of liver. The level of bilirubin in the paracetamol induced group (5.11 ± 0.66 mg/dl) was found greater compared to the control group (0.78 ± 0.29 mg/dl). This significant ($P< 0.001$) increase in bilirubin might be due to the damages caused in liver tissue. Silymarin decreased the bilirubin levels (1.19 ± 0.53 mg/dl) to near normalcy and also the ethanolic extract of *C. annuum* did the same. The low dose significantly ($P< 0.01$) brought back the bilirubin levels to (4.22 ± 0.48 mg/dl) and the high dose significantly brought ($P< 0.001$) back the values to (2.98 ± 0.53 mg/dl) which were in a dose dependent fashion (Table.4).

Table 4: Concentration of protein, albumin and total bilirubin in serum of control and experimental groups.

Groups	Total protein mg/dl	Albumin mg/dl	Total bilirubin mg/dl
Control	7.88 ± 0.52	4.20 ± 0.07	0.78 ± 0.29
Induced	4.67 ± 0.42###	2.74 ± 0.14###	5.11 ± 0.66###
Standard (Silymarin - 50 mg/kg b.w.)	8.03 ± 0.44***	4.34 ± 0.14***	1.19 ± 0.53***
Low dose (250 mg/kg b.w.)	7.32 ± 0.60***	4.44 ± 0.18***	4.22 ± 0.48**
High dose (500 mg/kg b.w.)	7.92 ± 0.35***	4.47 ± 0.16***	2.98 ± 0.53***

#Change in activities at $P<0.05$ when induced compared to control, ## $P<0.01$, ### $P<0.001$; *Change in activities at $P<0.05$ when standard, low dose and high dose compared to induced, ** $P<0.01$, *** $P<0.001$; Values are expressed as mean ± SD (n=6).

3.7 Effect of ethanolic extract of *C. annuum* red variety on serum liver markers

The liver function was assessed by determining the enzymes like AST, ALT and ALP which were secreted by the liver cells whose results are given in table 7 and figure 10. The induction of paracetamol resulted in a significant increase ($P< 0.001$) in the activities of all these enzymes in the serum which indicates the cellular damages in the liver. The activities of AST, ALT and ALP were found to be 154.33 ± 3.15 IU/L, 70.81 ± 1.97 IU/L and 143.28 ± 0.95 IU/L in the paracetamol group and 65.67 ± 1.49IU/L, 36.57 ± 2.00IU/L and 68.55 ± 0.68IU/L in the control group. The high dose, 400 mg/kg b.w. of the ethanolic extract of *C. annuum* significantly ($P< 0.001$) reduced the activities of these enzymes in the serum to 85.94 ± 1.75IU/L, 37.35 ± 1.86IU/L and 84.61 ± 2.18IU/L respectively. The activity of the ethanolic extract of *C. annuum* was similar to that of the standard drug, silymarin (Table 5).

Table 5: Concentration of serum liver markers (AST, ALT, ALP) of control and experimental groups.

Groups	AST IU/L	ALT IU/L	ALP IU/L
Control	65.67 ± 1.49	36.57 ± 2.00	68.55 ± 0.68
Induced	154.33±3.15###	70.81±1.97###	143.28±0.95###
Standard (Silymarin - 50 mg/kg b.w.)	75.95 ± 1.97***	37.35±3.42***	75.03 ± 0.81***
Low dose (250 mg/kg b.w.)	107.09±3.23***	56.45±2.60***	116.62±2.02***
High dose (500 mg/kg b.w.)	85.94 ± 1.75***	37.35±1.86***	84.61 ± 2.18***

#Change in activities at $P<0.05$ when induced compared to control, ## $P<0.01$, ### $P<0.001$; *Change in activities at $P<0.05$ when standard, low dose and high dose compared to induced, ** $P<0.01$, *** $P<0.001$; Values are expressed as mean±SD (n=6). [AST- Aspartate aminotransferase, ALT- Alanine aminotransferase, ALP- Alkaline Phosphatase]

4.0 DISCUSSION

Ripe fruits of red pepper (*Capsicum annuum* L.) are widely consumed as vegetables and are used as food colorants because they are a good source of the red carotenoids capsanthin and capsorubin. Capsanthin accounts for 30–60% of total carotenoids in fully ripe fruits (Minquez-Mosquera and Hornero-Mendez, 1994). It contains 11 conjugated double bonds, a conjugated keto group, and a cyclopentane ring, and has stronger antioxidative effects than β-carotene (Chen *et al*., 1996). These structural characteristics give rise to singlet oxygen-quenching ability (Hirayama *et al.,* 1994) and prevent colon carcinogenesis (Narisawa *et al*., 2000). In the present study pigments like lycopene and anthocyanin were quantified in the red bell peppers. The fresh fruits of *C. annuum* had appreciable amounts of both the pigments. These results justify that the *C. annuum* red variety could serve as best antioxidant. Earlier reports given by Kim *et al.,* (2002) suggests that pigment obtained from red bell peppers possess potent antioxidant activity.

Carbon tetrachloride is known to induce a significant increase in liver weight which may be due to blocking of secretion of hepatic triglycerides in to the plasma (Yoko *et al*., 2005). Similar increase in liver weight was observed in the present for the induction of paracetamol. Our results were in accordance with earlier reports of Anbarasu *et al*., (2011) and Sharma *et al*., (2012) who also say that there was a drastic increase in liver weight of paracetamol induced animals.

Paracetamol induction resulted in a decrease in glycogen storage ability of the liver. Similar decrease in glycogen levels was reported by Gautam *et al.,* (2012). The sustained depletion in glycogen may be due to reasons like, impaired mitochondrial respiration which may lower glycogen amounts due to enhanced glycolysis and low glutathione amounts which decrease the activity of enzymes involved in glycogen metabolism (e.g. glycogen synthase), thus lowering glycogen amounts (Gautam *et al.,* 2012). .

During toxicity, lipid profile of the serum and tissues increases. Intoxication of experimental animals with paracetamol altered the membrane structure and functions as shown by the increase in phospholipids, triglycerides and free fatty acids. Pretreatment with ethanolic extract of *C. annuum* decreased the lipid levels in tissues which might be due to alterations in the membrane fluidity. Earlier reports say that *Aphanizomenonflos-aquae* ethanolic extract also reduced the lipid levels in tissues by altering the membrane fluidity of the liver cells (Kuriakose and Kurup, 2010).

All the three glycoproteins namely hexose, hexosamine and sialic acid were found in decreased levels in the paracetamol induced group compared to control group. This drop in glycoprotein levels might be due to the depletion in the nucleotides which impairs the synthesis of protein and glycoprotein which leads to progressive damages to the cell membranes (Alqasoumi and Abdel-Kader, 2012).

Protein metabolism is a major project of liver and a healthy functioning liver is required for the synthesis of the serum proteins except for the γ globulins. Hypoproteinemia is a feature of liver damage due to significant fall in protein synthesis. Albumin is decreased in chronic liver disease and is generally accompanied by an increase in the β and γ globulins as a result of production of IgG and IgM (Kaplan *et al*., 1996). Hypoproteinemia was observed after paracetamol ingestion but the trend turns towards normal after *C.*

annuum treatment. Similar results were reported by Kanchana and Sadiq, (2011) and Venkatalakshmi *et al*., (2011).

Bilirubin is one of the most useful clinical clues to the severity of necrosis and its accumulation is a measure of binding, conjugation and excretory capacity of hepatocyte. Administration of paracetamol caused a significant elevation of bilirubin level has been attributed to the damage in the structural integrity of liver, because they are cytoplasmic in location and released into circulation after cellular damages indicating development of hepatotoxicity. Decrease in serum bilirubin after treatment with extract in liver damage induced by paracetamol, indicated the effectiveness of the extract in normal functional status of the liver (Manokaran *et al*., 2008).

Assessment of liver function can be made by the estimation of serum levels of metabolic enzymes like ALT, AST and ALP which are leaked out into systemic circulation during necrotic cell damage and hence are referred as sensitive indicators of liver injury (Molander *et al*., 1955; Nkosi *et al*., 2005). In present study, Paracetamol intoxication caused significant increase in these hepatic enzymes and this was probably due to the consequences of increased oxidative stress and necrotic cell death (Kyle *et al*., 1987). When there is heaptopathy, these enzymes leak into the blood stream in conformity with the extent of liver damage (Nkosi *et al.*, 2005). The elevated level of these entire marker enzymes observed in the group II, paracetamol treated rats in this present study corresponded to the extensive liver damage induced by toxin. The reduced concentrations of ALT, AST and ALP as a result of plant extract administration observed, during the present study, might probably be due in part to the presence of flavonoids.

Peppers contain moderate to high levels of neutral phenolics or flavonoids, phytochemicals that are important antioxidant components of a plant-based diet, other than traditional nutrients, that may reduce the risk of degenerative diseases (Hasler, 1998). The ascorbic acid, flavonoid, carotenoid, and total phenolic contents of peppers from three *C. annuum* species have already been reported (Howard *et al.,* 2000). Pepper fruits contain complex phenolic compounds, which occur with sugars as glycosides. Nevertheless, glycosides are extensively metabolized *in vivo* and the bioactive forms are not those found in plants. These reports justify the presence of secondary metabolites like flavonoid, carotenoid, and total phenolic compounds in *C. annuum*. A number of scientific reports indicated that certain flavonoids, triterpenoids and steroids have protective effect on liver due to its antioxidant properties. So the hepatoprotective effect rendered by the ethanolic extract of *C. annuum* could be attributed to the presence of flavonoid and phenolic compounds.

5.0 CONCLUSION

In present investigation it was found that ethanolic extract of *C. annuum* brought all the parameters affected by paracetamol toxicity near to normal. Thus the extract of *C. annuum* has hepatoprotective effect which minimizes the hepatotoxicity induced by paracetamol, thereby suggesting its use as a potent hepatoprotective agent.

6.0 REFERENCES

1. Russmann S, Gerd A, and Grattagliano I. Current Concepts of Mechanisms in Drug-Induced Hepatotoxicity. Current Medical Chemistry, 2009; 16: 3041-3053.
2. Vermeulen NPE, Bessems JGM, and Vandestreat R. Drug Metabolism Rev, 1992; 24: 367-407.
3. Ajith TA, Janardhanan KK. Antioxidant and anti hepatotoxic activities of Phellinusrimosus (Berk) Pilat. J. of Ethnopharmacol, 2002; 81: 387-391.
4. WHO, Regional Office for the Western Pacific, Research guidelines for evaluating the safety and efficacy of the herbal medicines. Manila. WHO, 1993.
5. Kren V, Walterova D. Silybin and Silymarin-new effects and applications. Biomed, 2005.
6. Ahmed M. Aboul-Enein1, Faten Abu El-Ela1, Emad A. Shalaby1 and Hany A. El-Shemy. Traditional medicinal plants research in Egypt: Studies of antioxidant and anticancer activities. J. Of Medicinal Plants Research, 2012; 6(5): 689-703.
7. Lakshmi Srinivas, Mohana Lakshmi S, NeelufarShama G, KoteswaraReddy K R, Prasanna. Medicinal Plants as Anti-Ulcer Agents. J. of Pharmacognosy and Phytochemistry, 2013; 2 (4): 91-97.
8. Giusti MM, Wrolstad, RE, Anthocyanins. Characterization and measurement with UV visible spectroscopy. Current Protocols in Food Analytical Chemistry, 2001.
9. Sadasivam S, Manickam A. Biochemical methods, 3rd edition. New Age International Publishers, 2009; 199-200.
10. Araya H, Horie T, Hayashi M, Awazu S. An alteration in liver microsomal membrane of the rat following paracetamol overdose. J.of Pharm Pharmacol, 1987; 39: 1047-49.
11. Folch J, Less M, and Solane SGH. A Simple method for the isolation and purification of total lipids from animal tissues. J. of Biological Chemistry, 1957; 226: 497-509.
12. Hron WT, and Menahan LA. A sensitive method for the determination of free fatty acids in plasma. J. of Lipid. Res, 1981; 22: 377-381.
13. Rice E C, Triglycerides in serum: Standard methods of clinical chemistry. Academic press: New York, 1970; 5: 215-222.
14. Rouser G, Fleischer S, and Yamamoto. Two dimensional thin layer chromatography separation of polar lipid and determination of phospholipids by

phosphorous analysis of spots. Lipids, 1970; 5: 494-6
15. Pari L, Karthikesan K. Protective role of tetrahydrocurcumin and chlorogenic acid on glycoprotein changes in streptozotocin-nicotinamide-induced diabetic rats. J. of Pharm. Sci. & Res, 2009; 1(4): 173-180.
16. Niebes P, Determination of enzymes and degradation products of glycosaminoglycans metabolism in the serum of healthy and varicose subjects. Clinical Chemistry Acta, 1972; 42: 399–408.
17. Wagner WD. A more sensitive assay discriminating galactosamine and glucosamine in mixtures. *Analytical Biochemistry*, 1979; 94(2): 394–396.
18. Maiti R, Jana D, Ghosh D. Antidiabetic effect of aqueous extract of seed of *Tamarindusindica*in streptozotocin induced diabetic rats. J. of Ethnopharmacol, 2004; 92: 85-91.
19. Minquez-Mosquera MI, and Hornero-Mendez D. Formation andTranformation of Pigments during the Fruit Ripening of*Capsicumannuum*cv. Bola and Agridulce. J. Agriculture FoodChemistry, 1994; 42: 38–44.
20. Chen Y, Nithianantham K, Shyamala M, Yoga Latha L, Subramanion LJ, SrSasidharan, S. Hepatoprotective potential of *Clitoriaternatea* leaf extract against paracetamol induced damage in mice. *Molecules*, 2011; 16: 10134-10145.
21. Hirayama O, Nakamura K, Hamada S, and Kobayasi Y. Singlet Oxygen-Quenching Ability of Naturally Occurring Carotenoids. Lipids, 1994; 29: 149–150.
22. Kim, Jaebok Park, Hwang. Changes in FA Composition and Antioxidative Activity of Pigment Extracts from Korean Pepper Powder (*Capsicum annuum* L.)Due to processing conditions. JAOCS, 2002; 79: 12.
23. Yoko A, Koyama T, Miyagic C, Inomata C, Kinoshita S, Ichiba T. Free Radical Scavenging and hepatoprotective action of the medicinal herb,*Crassocephalumcrepietioides* from Okinowa Islands. Biology Pharma Bull, 2005; 28.
24. Anbarasu C, BalasubramaniumRajkapoor, JayapaluKalpana. Protective effect of *PisoniaAculeata* on Paracetamol induced Hepatotoxicity in rats. J.of Experimental and Integrative Medicine, 2011; 1(3): 167-172.
25. Sharma, Sangameswaran, Mahajan SC, Manmeet Singh Saluja. Hepatoprotective Activity of Adina Cordifolia against Paracetamol induced Hepatotoxicity in rats. Asian Pacific J of Tropical Biomedicine, 2012; 1: 1-5.
26. Gautam R, Bhagawat Chandrasekar, Mukta Deobagkar-Lele, Srabanti Rakshit, Vinay Kumar B, Siva Umapathy, Dipankar Nandi. Identification of Early Biomarkers during Acetaminophen-Induced Hepatotoxicity by Fourier Transform Infrared Microspectroscopy. Plosone, 2012; 7(9): 1-13.
27. Kuriakose C, Kurup G. Antioxidant and hepatoprotective activity of Aphanizomenonflos-aquae Linn against paracetamol intoxication in rats. I. J. of Experimental Biology, 2010; 48: 1123-1130.
28. Alqasoumi I, and Abdel-Kader S. Screening of some Traditionally Used Plants for Their Hepatoprotective Effect, Phytochemicals as Nutraceuticals. Global Approaches to Their Role in Nutrition and Health, 2012; 255-278.
29. Kanchana N, and Mohamed Sadiq A. Hepatoprotective effect of *plumbagozeylanica*on paracetamol induced livertoxicity in rats. Int J.of Pharmacy and Pharmaceutical Sciences, 2011; 3: 151-154.
30. Manokaran S, Jaswanth S, Nandhakumar J, Duraisamy R, and Mallegaswari R. Hepatoprotective Activity of *Aervalanta* Linn against Paracetamol induced Hepatotoxicity in rats. Research J. of Pharma. And Technology, 2008; 1(4): 398-400.
31. Molander DW, Wroblewski F, La JSD. Transaminase compared with cholinesterase and alkaline phosphatase an index of hepatocellular integrity. Clinical Research. Proceedings, 1955; 3: 20-24.
32. Kyle, ME, Miccadei S, Nakae D. Superoxide dismutase and catalase protect cultured hepatocytes from the cytotoxicity of acetaminophen. Biochemical and Biophysics Research Communication, 1987; 149: 889-896.
33. Nkosi CZ, Opoku AR, Terblanche SE. Effect of pumpkin seed (*Cucurbitapepo*) protein isolate on the activity levels of certain plasma enzymes in CCl4-induced liver injury in lowprotein fed rats. Phytotherapy research, 2005; 19: 341-345.
34. Howard LR, Talcott ST, Brenes CH, and Villalon B. Changes in Phytochemical and Antioxidant Activity of Selected Pepper Cultivars (Capsicum annuum) As Influenced by Maturity. Journal of Agriculture, 2000; 48: 1713-1720.