INTERNATIONAL RESEARCH JOURNAL OF PHARMACY
www.irjponline.com
ISSN 2230 – 8407

Research Article

HEPATOPROTECTIVE EFFECT OF GOTU KOLA (*CENTELLA ASIATICA* LINN.) ON CARBON TETRA CHLORIDE INDUCED LIVER INJURY IN RATS

Smitha M. Xavier[1]* and D. Umadevi[2]
[1]Rubber Research Institute of India, Kottayam, Kerala, India
[2]Department of Zoology, University of Calicut, Kerala, India
*Corresponding Author Email: smitha.rrii@yahoo.com

Article Received on: 27/09/14 Revised on: 11/11/14 Approved for publication: 26/11/14

DOI: 10.7897/2230-8407.0512189

ABSTRACT
Centella asiatica Linn. is a well known medicinal herb used in various types of diseases. The present study was conducted to evaluate the hepatoprotective effects of the *Centella asiatica* extract in carbon tetrachloride (CCl_4) induced liver injury in rats. Biochemical parameters such as serum total protein, albumin and marker enzymes (aspartate aminotransferase, alanine aminotransferase and alkaline phosphatase) were estimated both before and after the experiment. The present investigation reports that *Centella asiatica* affords protection against $CC1_4$ induced liver injury as it restored marker enzyme levels, total protein and albumin levels compared to that of untreated control.
Keywords: *Centella asiatica* Linn., hepatoprotective, ayurveda, carbon tetrachloride.

INTRODUCTION

Centella asiatica Linn. Belonging to family of Umbeliferae popularly known as "Brahmi", is a very useful medicinal plant described by Charaka as an anti aging plant. In Ayurveda it has been used in the treatment of skin diseases, local wounds and also for improving general mental ability, jaundice and hepatitis[1,2]. It is mainly used to treat neurological disturbances[3]. Apart from its role as a brain tonic, it has been reported to have activities in wound healing[4,5], anti-inflammatory[6,7], antioxidant[8,9], antibacterial[10], antifungal[11] and antidiabetic[12] treatments. Chemical studies on *Centella asiatica* leaves showed that one of the major components is asiaticoside[13]. The present investigation is an attempt to evaluate the short term protective effect of *Centella asiatica* in carbon tetrachloride induced hepatic damage.

MATERIALS AND METHODS

Preparation of the extract of *Centella asiatica*

The medicinal plant used in this experiment is *Centella asiatica.* The plant was collected from Angamaly, Ernakulam, Kerala, India. The plant is found in moist soil, especially along bunds and canals. The crude extract was prepared by grinding the whole plant. 1 g of crude extract was dissolved in 25 ml of purified water. It was given at a dosage of 200 mg/kg body weight.

Animal model of CCl_4-induced liver injury

Animals were purchased from Small Animals Breeding Station (SABS), Veterinary College, Mannuthy, Thrissur, Kerala, India. The study was conducted at Little Flower Medical Research Centre, Angamaly, after obtaining clearance from Institutional Animal Ethics Committee. The animals were kept at room temperature of 26-28^0 C and humidity of 56-58 %. They were fed with standard diet (prepared in the lab) and water *ad libitum*. The animals were kept for two weeks for acclimatization before starting the experiment. Female rats of *Sprague dawley* weighing 180-280 g were selected for the experiment. The animals were divided into three groups of six animals each. Experimental groups were induced hepatic damage, by intra peritoneal injection of 1:1 mixture of $CC1_4$ and coconut oil, 1 mg/kg body weight, thrice in a week. Group A served as control (1 ml distilled water /100 g body weight), Group B served as Untreated control (1 ml $CC1_4$ – coconut oil mixture/kg body weight intraperitoneally + 1 ml distilled water orally) and Group C served as Treated group (1 ml $CC1_4$ – coconut oil mixture/kg body weight intraperitoneally + 200 mg/kg of extract orally). The treatment was given for a period of seven days.

Estimation of biochemical parameters

Blood samples were collected from caudal vein of rats under ether anesthesia for the estimation of biochemical parameters such as Alanine transaminase (ALT), Aspartate transaminase (AST), Alkaline Phosphatase (ALP), total protein and albumin. Estimation of serum aminotransferases were done by 2,4-DNPH method[14]. Alkaline phosphatase was estimated by Kind and King method[15]. Serum total protein was estimated by Biuret method[16] and albumin estimation was done by Bromocresol green (BCG) method[17].

RESULTS

Effect of *Centella asiatica* on liver function tests

The level of serum ALP in the control group (A) was in the normal range. The serum ALP increased to a significant level ($p < 0.001$) in untreated control (B) compared to normal group. The percentage of increase observed in group (B) was 58 %, while there was only 16 % increase in the treatment group (C). A significant decrease ($p < 0.001$) in ALP (Table 1) was observed in the treated group compared to untreated control group. There was 36 % decrease in the treatment group (C). The amount of ALT showed a remarkable reduction in the extract treated group when compared untreated group. The level of serum in the AST in the control group (A) appeared in the normal range. The serum AST increased to a significant level ($p < 0.001$) in untreated control (B) compared to normal group. The percentage of

increase observed in group (B) was 96 %, while there was only 29 % increase in treatment group (C). A significant decrease ($p < 0.001$) in AST level (Table 1) was observed in the treated group compared to untreated control group. There was 52 % decrease in the treatment group (C). The level of serum albumin in the control group (A) was in the normal range. The serum albumin decreased to a significant level ($P < 0.001$) in untreated control (B) compared to normal group. The percentage of decrease observed in group (B) was 49 % while there was only 35 % decrease in the treatment group (C). A significant increase ($P < 0.001$) albumin level (Table 2) was observed in the treated group compared to untreated control group. There was 11 % increase in the treatment group (C). The level of serum total protein in the control group (A) was in the normal range. The serum total protein decreased to a significant level ($p < 0.001$) in untreated control (B) compared to normal group. The percentage of decrease observed in group (B) was 37 %, while there was only 18 % decrease in the treatment group (C). A significant increase ($p < 0.001$) increase in protein level (Table 2) was observed in the treated group compared to untreated control group. There was 16 % increase in the treatment group (C). The above effects on the total protein, albumin and liver marker enzymes revealed that crude extract of *Centella asiatica* restored liver functions to an almost normal level, compared to untreated control group.

Effect of *Centella asiatica* extract on liver

Microscopic study

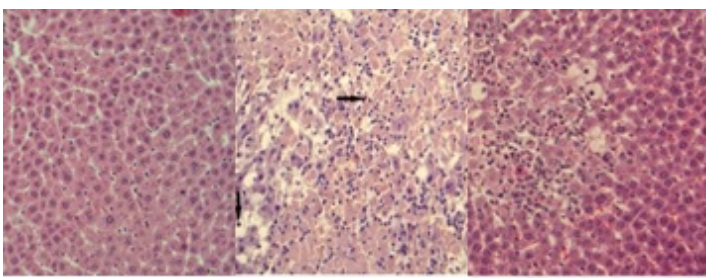
Animals were sacrificed at the end of seven days study. Microscopic studies of the liver showed normal liver in reddish brown color in control group (A). The liver of untreated control animal, group (B) was in pale color. In the treatment group (C), liver appeared to be normal as in the control group (A).

Table 1: Effect of *Centella asiatica* on ALP, ALT and AST in CCl_4 induced hepatic damage

No	Alkaline phosphatase (IU/L)			Alanine Transamiase (IU/L)			Aspartate Transamiase (IU/L)		
	Control (A)	Untreated Control (B)	Treated animal (C)	Control (A)	Untreated Control (B)	Treated animal (C)	Control (A)	Untreated Control (B)	Treated animal (C)
1	38	59	42	18	40	36	22	46	30
2	37	58	46	22	42	28	21	39	26
3	39	59	41	20	44	30	20	41	26
4	36	62	47	19	45	29	23	42	27
5	38	61	43	21	43	29	22	42	28
6	39	60	45	20	42	33	21	43	29
Mean	37.83	59.8	44	20	42.66	30.83	21.5	42.17	27.67
S.D	1.169	1.472	2.366	1.414	1.751	3.061	1.049	2.317	3.011

Table 2: Effect of *Centella asiatica* on albumin and total protein in CCl_4 induced hepatic damage

No	Albumin (g/dl)			Total protein (g/dl)		
	Control (A)	Untreated Control (B)	Treated animal (C)	Control (A)	Untreated Control (B)	Treated animal (C)
1	3.4	2.1	2.4	6.1	4.9	5.5
2	3.6	2.2	2.5	7.0	4.9	5.4
3	3.5	2.4	2.6	6.2	4.5	5.5
4	3.4	2.5	2.4	6.5	4.6	5.5
5	3.4	2.3	2.8	6.4	4.7	5.5
6	3.5	2.4	2.7	6.6	4.8	5.4
Mean	3.46	2.32	2.57	6.47	4.73	5.47
S.D	0.0817	0.1472	0.1633	0.3204	0.1633	0.05164



A B C

Figure 1: **Effects of *Centella asiatica* pretreatment on acute liver damage induced by CCl_4 treatment in the rats**

Microscopic studies of the liver showed normal liver in reddish brown color in control group (A). The liver of untreated control animal was in pale color group (B). In the treatment group (C), liver appeared to be normal as in the control group

DISCUSSION

Intra peritoneal injection of carbon tetrachloride induces toxicity in liver. $CC1_4$ is a simple molecule, which when administered, cause centriolobular hepatic necrosis and fatty liver. Low doses of $CC1_4$ produce only fatty liver and destruction of hepatic cytochrome P-450. However chronic administration or exposure leads to liver cirrhosis, in some instance liver cancer and kidney damage[18]. Hepatic cells are normally protected from injury with glutathione content of hepatocytes available for detoxification. When glutathione gets exhausted hepatocytes become vulnerable to noxious effects of metabolites resulting in necrosis of liver[19] $CC1_4$ is metabolized in the liver to toxic free radical $CC1_3$ which affect cellular permeability of hepatocytes, leading to elevated levels of serum biochemical parameters like SGOT, SGPT and ALP[20]. The serum total protein and albumin levels were decreased and the serum marker enzyme levels were increased during $CC1_4$ induced hepatic damage. After seven days treatment with *Centalla asiatica* extract, the level of total protein and albumin were increased and reached to almost normal values as in the control animals. The level of marker enzymes ALP, ALT and AST were decreased to normal levels as in the control animals. It is concluded that *Centella asiatica* affords protection against $CC1_4$ induced liver injury as it restored marker enzyme levels, total protein and albumin levels compared to that of untreated control. Earlier studies show that the extract of the plant prevented fatty infiltration of liver in rats and caused restoration of 40 % of damaged liver cells to the normal state. The present study examined the hepatocurative activity of *Centella asiatica* using carbon tetrachloride induced liver injury in rats. The activities of amino transferases and alkaline phosphatase were elevated and level of total protein and albumin were reduced in carbon tetrachloride induced rats. *Centella asiatica* extract was administered orally at 200 mg/kg/day for seven days. After the treatment period, the level of aminotransferases and alkaline phosphatase were reduced. While that of the total protein and albumin were increased near to normal level as in the control animals. Hence the hepatocurative effect produced by *Centella asiatica* in carbon tetrachloride induced liver injury is attributed to its potent antioxidant action, particularly to the asiaticoside present, which in turn inhibits the hepatic damage caused by carbontetrachloride through free radical mediator reactions.

ACKNOWLEDGEMENT

We thank Little Flower Medical Research Centre, Angamaly, Kerala, India for providing laboratory facilities for this research work.

REFERENCES

1. Arora D, Kumar M and Dubey SD. *Centella asiatica* - A review of its medicinal uses and pharmacological effects. Journal of Natural Remedies 2002; 2 (2): 143-149.
2. Kashmira Gohil J, Jagruti Patel A and Anuradha Gajjar K. Pharmacological Review on *Centella asiatica*: A Potential Herbal Cure-all. Indian Journal of Pharmaceutical Sciences 2010; 72(5): 546–556. http://dx.doi.org/10.4103/0250-474X.78519
3. Ilkay Erdogan Orhan. *Centella asiatica* (L.) Urban: From Traditional Medicine to Modern Medicine with Neuroprotective Potential. Evidence-Based Complementary and Alternative Medicine 2012; 1-8.
4. Tenni R, Zanaboni G, De Agostini MP, Rossi A, Bendotti C and G Cetta. Effect of the triterpenoid fraction of *Centella asiatica* on macromolecules of the connective matrix in human skin fibroblast cultures, Italian Journal of Biochemistry 1988; 37(2): 69–77.
5. Suguna L, Sivakumar P and Chandrakasan G. Effects of *Centella asiatica* extract on dermal wound healing in rats. Indian Journal of Experimental Biology 1996; 34(12): 1208–1211.
6. Somchit MN, Sulaiman MR, Zuraini A *et al*. Antinociceptive and anti inflammatory effects of *Centella asiatica*. Indian Journal of Pharmacology 2004; 36(6): 377–380.
7. George M, Joseph L and Ramaswamy. Anti-allergic, anti-pruritic, and anti-inflammatory activities of *Centella asiatica* extracts. African Journal of Traditional, Complementary and Alternative Medicines 2009; 6(4): 554–559.
8. Jayashree G, Kurup Muraleedhara G, Sudarslal S and Jacob VB. Anti-oxidant activity of *Centella asiatica* on lymphoma-bearing mice. Fitoterapia 2003; 74(5): 431–434. http://dx.doi.org/10.1016/S0367-326X(03)00121-7
9. Bajpai M, Pande A, Tewari SK and Prakash D. Phenolic contents and antioxidant activity of some food and medicinal plants. International Journal of Food Sciences and Nutrition 2005; 56(4): 287–291. http://dx.doi.org/10.1080/09637480500146606
10. Zaidan MR, Noor Rain A, Badrul AR, Adlin A, Norazah A and Zakiah I. *In vitro* screening of five local medicinal plants for antibacterial activity using disc diffusion method. Tropical Biomedicine 2005; 22(2): 165–170.
11. Naz E and Ahmad M. Evaluation of five indigenous medicinal plants of Sindh, Pakistan for their antifungal potential. Pakistan Journal of Scientific and Industrial Research 2009; 52(6): 328–333.
12. Venu Gopal Rao ML and Mastan SA. Antidiabetic effects of methanolic extract of *Centella asiatica* (Linn.) on induced hyperglycemic rats. Biosciences Biotechnology Research Asia 2007; 4(2): 721–724.
13. Bonte F, Dumas M, Chaudagne C and Meybeck A. Influence of asiatic acid, madecassic acid, and asiaticoside on human collagen I synthesis. Planta Medica 1994; 60 (2): 133-135. http://dx.doi.org/10.1055/s-2006-959434
14. Kind PRN and King EJ. Estimation of plasma phosphatases by determination of hydrolyzed phenol with amino antipyrine. Journal of Clinical Pathology 1954; 7: 330-332. http://dx.doi.org/10.1136/jcp.7.4.322
15. Reitman S and Frankel S. A colorimetric method for the determination of serum glutamic oxaloacetic and glutamic pyruvate aminotransferases. American Journal of Clinical Pathology 1957; 28: 53-56.
16. Lowry OH, Rosehmugh NJ, Farr AL and Ra Dall KJ. Protein measurement with the Folin phenol reagent. Journal of Biological Chemistry 1951; 193: 265-275.
17. Ackermann P and Toro G. Practical clinical chemistry, 1st edition. Little Brown and Company, Boston, USA; 1975.
18. Farrell GC, Larter CZ. Nonalcoholic fatty liver disease: from steatosis to cirrhosis. Hepatology 2006; 43(2): S99-S112. http://dx.doi.org/10.1002/hep.20973
19. Vichitra Kaushik, Gagandeep Chaudhary, Shoaib Ahmad, Vipin Saini, A Pandurangan. Diverse Sources of Hepatotoxicity in rats – A review. International Journal of Pharmaceutical Sciences Review and Research 2013; 21(1): 254-259.
20. Jayasekar P, Mohanan PV, Rathinam K. Hepatoprotective activity of ethyl acetate extract of *Acacia catechu*. Indan Journal of Pharmacology 1997; 29: 426.

Cite this article as:

Smitha M. Xavier and D. Umadevi. Hepatoprotective effect of Gotu kola (Centella asiatica Linn.) on carbon tetra chloride induced liver injury in rats. Int. Res. J. Pharm. 2014; 5(12):929-931 http://dx.doi.org/10.7897/2230-8407.0512189

Source of support: Nil, Conflict of interest: None Declared