The Pharma Innovation Journal 2022; 11(7): 3324-3328

www.ThePharmaJournal.com

The Pharma Innovation

ISSN (E): 2277-7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2022; 11(7): 3324-3328

www.thepharmajournal.com
Received: 20-04-2022
Accepted: 08-06-2022

BH Choudhury
Department of Biochemistry and Agricultural Chemistry, SCSCA, AAU, Rangamati, Dhubri, Assam, India

S Islam
Department of AHD, B.N. College of Agriculture, Biswanat Chariali, AAU, Sonitpur, Assam, India

Om Singh
Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya Campus-College of Horticulture, Mandsaur, Madhya Pradesh, India

P Mohan
Department of Pharmacology and Toxicology, AAU, Khanapara, Guwahati, Assam, India

Corresponding Author:
BH Choudhury
Department of Biochemistry and Agricultural Chemistry, SCSCA, AAU, Rangamati, Dhubri, Assam, India

Biochemical evaluation and characterization of *Scorporia dulcis* L. against chemically induced jaundice in albino rats

BH Choudhury, S Islam, Om Singh and P Mohan

Abstract
The crude and purified extracts of *Scorporia dulcis* were used to study the hepatoprotective/ hepatocurative activity in albino rats. Two doses of extracts *viz*. 100mg, 300mg of crude and 10mg, 30mg of purified extracts per kg body weight were given orally to albino rats for 7 days. On the 3rd and 6th days the animal, received CCl_4 @3ml/kg body weight and olive oil subcutaneously (50:50). Control group of animals were also maintained. On the 8th day of the experiment, blood samples were collected by direct puncture of the heart and the serum was extracted from the samples. The animals were then sacrificed by chloroform and the tissue samples (liver) were used for hepatoprotective studies. To assess the extent of hepatic damage and to monitor the degree of hepatoprotection of the extracts serum alkaline phosphatase, serum-γ-glutamyl transferase, serum glutamate pyruvate transaminase, total bilirubins were assayed. It was observed that the animals fed with the crude and purified extracts exhibited statistically highly significant hepatoprotective action in comparison to animals treated with only CCl_4 which resulted hepatic disorder.

Keywords: *Scorporia dulcis*, Jaundice, crude extract, hetaoprotective, albino rats. CCl_4

Introduction
Liver diseases are a serious health problem. In the absence of reliable liver protective drugs in allopathic medical practice, herbs play a role in the management of various liver disorders. Numerous medicinal plants and their formulations are used for liver disorders in ethanomedical practices and in traditional system of medicine in India. However, we do not have satisfactory remedy for serious liver disorder; most of the herbal drugs speed up the natural healing process of liver. Recently interest in medicinal plants research increased all over the world. It has been reported that medicinal plants, used in various traditional systems, have immune potential against various diseases [1]. More than 13,000 plants have been studied during last five years for various pharmacological properties. Indian medicinal plants belonging to about 40 families were investigated as liver protective drugs [2]. Plant drugs are widely used in the Northeast part of India for the cure of various diseases including jaundice and other liver disorders [3]. The extract of whole plant of *Scorporia dulcis* are widely used in N.E. region for the treatment of jaundice since long [4]. Liver is the organ which is most susceptible to the toxic effects of carbon tetra chloride (CCl_4) due to overdoses. The present study is aimed to evaluate hepatoprotective/hepatocurative effects of *Scorporia dulcis* on CCl_4 induced liver damage in albino rats.

Materials and Methods
The whole plant of *Scorporia dulcis* was collected by uprooting the plant from Jorhat district, Assam. The samples after cleaning were dried in shade, powdered in cutter mill (IKON instrument, Delhi) and extracted with distilled methanol. The crude extract of *Scorporia dulcis* was prepared by dissolving the cleaned, dried powder of whole plant in distilled methanol for 48 hours at room temperature. The purified extracts were prepared by loading10g of dried crude extract in a column of silica gel 300g placed with Hexane and 400 ml of fraction-1(hexane), fraction-2(Ethyl acetate/hexane) and fraction-3(Chloroform) descended bellow were collected. These extracts were evaporated in rotary evaporator (Bibby, Sterilin) under reduced pressure. The viscous semi-liquid extracts obtained from rotary evaporator were dried in lyophilizer (Heto, LYO LAB3000) at -55ºC. Methanol and CCl_4 were obtained from Ranbaxy Laboratories.

Sprague Dawley albino rats were procured locally and bred in Department of Pharmacology and Toxicology, College of Veterinary Science, Assam Agricultural University, Khanapara, Guwahati, Assam with the approval of appropriate ethical committee of the College. The animals were housed in clean polypropylene cages and given standard ration with ad-libitum supply of drinking water. Twenty four rats of either sex weighing between 130-160 gm were chosen and were divided into four groups containing six rats in each group.

The treatment protocol was planned to study the role of crude and purified extracts of *Scorporia dulcis* in both preventive and curative aspects of CCl_4 induced hepatotoxicity.

GroupI:(Vehicle control) Received tween 80 (20%) orally for seven days and on 3rd and 6th days received normal saline and olive oil subcutaneously.

GroupII: (CCl_4 treated group) received tween 80 (20%) orally for seven days and on 3rd and 6th days received CCl4 @3ml per kg body weight and olive oil subcutaneously.

Group III: Received crude extract @ 100mg and purified extract of *Scorporia dulcis* @10mg per kg body weight along with tween 80 (20%) as vehicle orally for seven days and on 3rd and 6th days received CCl4 @3ml per kg body weight and olive oil subcutaneously.

Group IV: Received crude extract @ 300mg and purified extract of *Scorporia dulcis* @30mg per kg body weight along with tween 80 (20%) as vehicle orally for seven days and on 3rd and 6th days received CCl4 @3ml per kg body weight and olive oil subcutaneously.

The treatment protocol was planned to study the hepatoprotective effect of *Scorporia dulcis* on CCl_4 induced hepatotoxicity**.**

Group-I, received tween 80 (20% tween 80 concentration was used as it was observed that the extracts of *Scorporia dulcis* was not soluble in saline or water but soluble in 20% concentration of tween 80) for 7 days and on the 3rd and 6th days, received normal saline and olive oil sub-cutaneously.

Group-II, received tween 80 (20%) for 7 days and on the 3rd and 6th days, received CCl_4, @ of 3ml per Kg. body weight with olive oil sub-cutaneously

Group-III & Group-IV, received100mg/ Kg body weight and 300mg/Kg body weight of crude extract of *Scorporia dulcis* orally with 20%Ttween 80 as vehicle, respectively, for 7 days, and on 6th day received CCl_4 @ of 3ml/ Kg body weight in olive oil subcutaneously.

Group-V, received tween 80 (20%) for 7 days and on the 3rd and 6th days, received normal saline and olive oil sub-cutaneously.

Group-VI, received tween 80(20%) for 7 days and on the 3rd and 6th days, received CCl_4, @ of 3ml per Kg. body weight, with olive oil sub-cutaneously

Group-VII, received10mg/ Kg body weight of Hexane extract of *Scorporia dulcis* orally, with Tween 80 as vehicle, for 7 days, and on 6th day received CCl_4 @ of 3ml/ Kg body weight in olive oil subcutaneously.

Group-VIII, received 30mg/ Kg body weight of Hexane extract of *Scorporia dulcis*, orally, with Tween 80 as vehicle, for 7 days, and on 6th day received CCl_4 @ of 3ml/ Kg body weight in olive oil subcutaneously.

Group-IX, received10mg/Kg body weight of Ethyl Acetate/.Hexane extract of *Scorporia dulcis*, orally, with Tween 80 as vehicle, for 7 days, and on 6th day received CCl_4 @ of 3ml/ Kg body weight in olive oil subcutaneously.

Group-X received 30mg/ Kg body weight of Ethyl Acetate/Hexane extract of *Scorporia dulcis*, orally, with Tween 80 as vehicle, for 7 days, and on 6th day received CCl_4 @ of 3ml/ Kg body weight in olive oil subcutaneously.

Group-XII, received10mg/Kg body weight of Chloroform extract of *Scorporia dulcis*, orally, with Tween 80 as vehicle, for 7 days, and on 6th day received CCl_4 @ of 3ml/ Kg body weight in olive oil subcutaneously.

Group-XII, received 30mg/ Kg body weight of Chloroform extract of *Scorporia dulcis*, orally, with Tween 80 as vehicle, for 7 days, and on 6th day received CCl_4 @ of 3ml/ Kg body weight in olive oil subcutaneously.

On the 8th day of the experiment blood samples were collected by direct puncture of the heart and serum was collected from the samples. The blood serum samples were collected for analysis of biochemical parameters. The activities of serum hepatic marker enzymes, namely serum alkaline phosphatase (SAP), serum-γ-glutamyl transferase (SGPT), serum glutamate pyruvate transaminase (SGOT), total bilirubin were assayed in serum using standard kits obtained from Ranbaxy Laboratories and PDP, Spawn. The results were expressed as international units/ liter (IU/L).The statistical analysis was carried out by one way Analysis of Variance (ANOVA)

Results & Discussion

In the present study, a dose of 3ml of CCl_4 per kg body weight was used to induce liver damage in the rat as indicated by a significant ($P< 0.05$, $P< 0.01$, $P< 0.001$) elevation of serum marker enzymes *viz*. SAP, SGPT, SGOT and a significant ($P< 0.01$, $P< 0.001$) reduction of serum total bilirubin, total protein (Table 1, 2, 3 and 4). The results are in agreement with the earlier reports [5, 6, 7]. Histopathological changes has been seen which confirmed the hepatic damages. Compared to normal liver tissue, carbon tetrachloride (CCl_4) treatment showed extensive centrilobular necrotic patches extending to midzone with neutrophilic collection. Central to central bridging necrosis was seen, however the liver tissue of rats fed with the purified extracts of *Scorporia dulcis* L significantly improved the necrotic symptoms (Figures 1, 2 &3).

Both preventive and curative aspects of the liver were studied. Liver has got the capacity to regenerate. This regenerative capacity was assessed by administering 3ml CCl_4 per kg body weight orally for two days and with a vehicle for seven days. The results as shown by group II indicate that no regeneration had taken place [8]. This group also acted as control for curative treatment. The extracts of *Scorporia dulcis* exhibited an ability to counteract the CCl_4 induced changes in the biochemical parameters both in preventive and curative measures.

Table 1: Effect of crude extracts of *Scorporia dulcis* on biochemical parameters in albino rats.

Group	SAP K.A. units	SGOT U/ml	SGPT U/ml	Bilirubin g/dl
1. GroupI(Control)	27.76±1.00	140.68±7.42	67.55±1.11	0.53±0.06
2. GroupII(CCl_4)	55.60±0.69**	290.83±4.15**	193.4±5.80**	1.50±0.11*
3.GroupIII(Preventive)	42.20±0.57*	204.67±4.74**	130.67±4.74*	0.70±0.12
4.GroupIV(Curative)	38.27± 0.37*	160.51±3.40**	115.60±3.09**	0.78±0.03*

*=P< 0.05 and **= P< 0.01

SAP= Serum alkaline phosphatase, SGOT= Serum-γ-Glutamyl Transferase, SGPT= Serum Glutamate Pyruvate Transaminase

Table 2: Effect of purified fraction-1 (Hexane) of crude extracts of *Scorporia dulcis* on biochemical parameters in albino rats.

Group	SAP K.A units	SGOT U/ml	SGPT U/ml	Bilirubin g/dl	Total protein g/dl
1.GroupI(Control)	39.90±3.10.	42.61±3.14	36.20±3.40	0.44±0.04	7.41±0.40
2.GroupII(CCl_4)	92.66±4.58**	296.13±20.74**	80.06±2.26**	1.30+0.08**	4.28±0.43**
3.GroupIII(Preventive)	72.42±3.06*	227.46±18.45	50.54±3.83**	0.91±0.02**	5.09±0.17
4.GroupIV(Curative)	78.34±2.94*	225.08±18.28	48.50±2.23**	0.83±0.04**	5.07±0.15

*=P< 0.05 and **= P< 0.01

Table 3: Effect of the purified fraction-2 (Ethyl Acetate/Hexane, 1:10) of crude extracts of *Scorporia dulcis* on biochemical parameters in albino rats.

Group	SAP K.A units	SGOT U/ml	SGPT U/ml	Bilirubin g/dl	Total protein g/dl
1.GroupI(Control)	35.98±3.12.	43.61±4.14	34.26±3.41	0.42±0.04	7.31±0.41
2.GroupII(CCl_4)	91.56±4.68**	291.13±22.74**	80.06±3.26**	1.34±0.08**	4.22±0.43**
3.GroupIII(Preventive)	59.18±4.33**	161.67±8.33**	41.0±7.34**	0.93±0.02**	5.15±0.09
1.GroupI(Control)	54.32±1.97**	211.33±8.84**	34.0±5.41**	0.85±0.04**	5.02±0.21

*=P< 0.05 and **= P< 0.01

Table 4: Effect of the purified fraction-3(Ethyl Acetate/Hexane: 1:7) of crude extracts extracts of *Scorporia dulcis* on biochemical parameters in albino rats.

Group	SAP K.A units	SGOT U/ml	SGPT U/ml	Bilirubin g/dl	Total protein g/dl
1.GroupI(Control)	36.98±3.12.	43.61±4.14	34.26±3.41	0.43±0.04	7.31±0.41
2.GroupII(CCl_4)	90.56±4.68**	292.13±22.74**	80.06±3.26**	1.30±0.08**	4.26±0.43**
3.GroupIII(Preventive)	72.80±4.07*	230.33±6.58	39.67±6.81**	0.81±0.05**	4.45±0.13
1.GroupI(Control)	69.61±4.67*	246.00±2.58**	36.33±6.23**	0.86±0.03**	4.78±0.10

*=P< 0.05 and **= P< 0.01

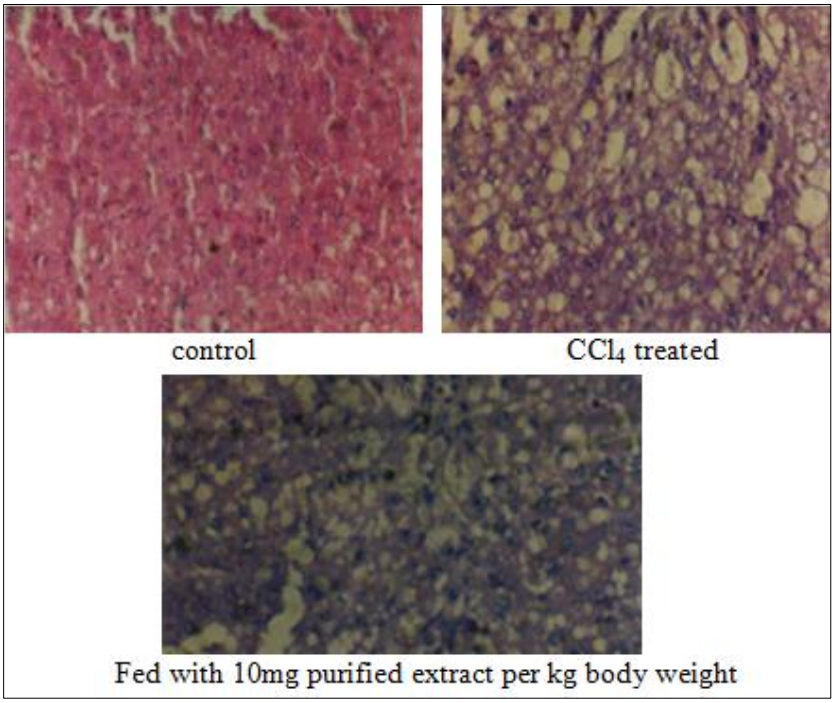

Fig 1: Histopathological changes of the liver tissues of albino rats fed with purified extract of *Scorporia dulcis*, fraction-1 (Hexane) (Magnification: 40x2.5).

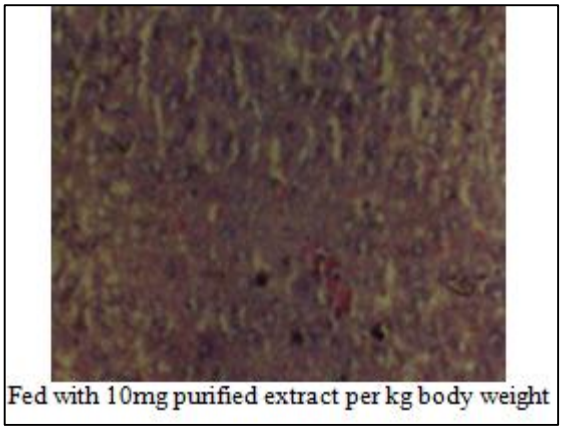

Fig 2: Histopathological changes of the liver tissues of albino rats fed with purified extract of *Scorporia dulcis*, fraction-2 (Ethyl Acetate+Hexane 1:7) (Magnification: 40x2.5).

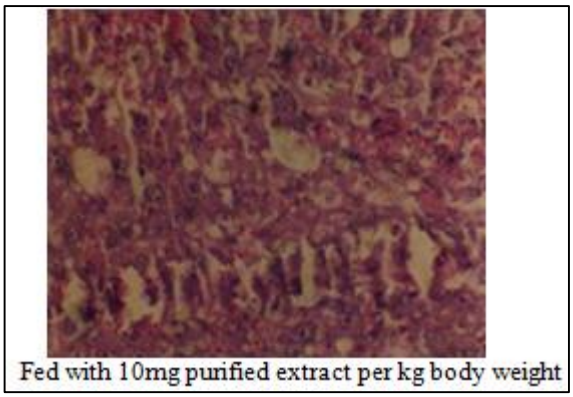

Fig 3: Histopathological changes of the liver tissues of albino rats fed with purified extract of *Scorporia dulcis*, fraction-3 (Ethyl Acetate+Hexane 1:10) (Magnification: 40x2.5).

Significant hepatoprotective efficacy has been reported earlier in various plants [9]. Liver cells participate in a variety of metabolic activities and thus contain a host of enzymes. In severe acute liver damage serum transaminases level parallel to those of organs indicating that both cellular and mitochondrial membranes have been damaged. It is reported that large doses of CCl_4 results in cell lysis and cytoplasmic hepatic enzymes are released into blood circulation [10]. Many fold increase of enzyme leakage as demonstrated by increased level serum enzymes ALT and AST has been noted indicating liver cell damage by CCl_4 [11]. It has also been supported by other workers [12]. Since membrane intrigrity is linked with intracellular metabolic states, disturbance in later results in membrane lesion with concomitant increase in enzyme leakage giving rise to hypoxia and membrane hypermeability.

The extracts of *Scorporia dulcis* are able to prevent hepatic injury/liver necrosis in preventive group and enhanced regeneration in curative groups [13]. This was evidenced by lowering the activity of Serum marker enzymes *viz*. Serum alkaline phosphatase, Serum-γ-Glutamyl Transferase, Serum Glutamate Pyruvate Transaminase and a significant reduction of serum total bilirubin. Elevation of activities of these enzymes in serum indicated membrane damage [14]. SAP and SGOT are better index of liver injury [15]. More than 50% reduction of activities of these enzymes indicated recovery of hepatocytes in spite of insult due to hepatotoxin by CCl_4. Hepatotoxicity might be due to lipid peroxidation, depletion of glutathione/cytochrome P-450, an altered immunological system induced by various chemical agents or direct damage to the cell [16, 17].

It can be concluded that the significant antihepatotoxic activity shown by extracts of *Scorporia dulcis* may due to its inhibitory effect on serum enzymes or lipid peroxidation. The plant extract may interfere with cytochrome P-450 and ultimately hinder the formation of hepatotoxic CCl_3 free radical and exert its hepatoprotective action. The extract may also have antioxidant property which inhibited the deleterious effect of free radicals generated by CCl_4 influencing the membrane rigidity by prevention or inhibition of membrane peroxidation [18]. Thus present study indicated that the *Scorporia dulcis* protect the liver damage against CCl_4 induced hepatotoxicity.

Acknowledgements

The authors are grateful to the ICAR, New Delhi for providing fund in the form of a research project.

References

1. Rates SMK. Plants as source of drugs. Toxicon. 2001;39:603.
2. Handa SS, Sharma A, Chakrabarty KK. Natural products and plant as liver protecting agents. Fitoterapia. 1986;57:307
3. Borah RC, Baruwa J, Choudhury BH, Baruah IC, Baruah NC. Chemical investigation of *Phyllanthus fraternus* and *Glycosmis pentaphylla*, Traditional Plants Used against Jaundice in the Bhramaputra Valley. In Bioprospecting of Commercially Important Plants In Bioprospecting of Commercially Important Plants, 2004, 282.
4. Gogoi P, Borthakur B. Traditional medicine used against jaundice by people of Brahmaputra Valley, Assam. India. Adv. Plant Sci. 1997;9(II):131.
5. Shenoy KA, Somaya SN, Bairy KL. Evaluation of hepatoprotective activity of *Ginko biloba* in rats. Indian J. Physiol Pharmacol. 2002;46(2):167.
6. Emmanuel S, Amalraj, Ignacimuthu S. Hepatoprotective effect of coumestains isolated from the leaves of *Wedelia calendulacea*. Indian Journal Exp. Biol. 2001;39:1305.
7. Singh B, Saxena AK, Chandan BK, Agarwal SG, Anand KK. *In vivo* hepatoprotective activity of active fraction from ethanolic extract of *Eclipta alba* leaves. Indian J. Physiol Pharmacol. 2001;45(4):435.
8. Asha VV. Preliminary studies on the hepatoprotective activities of *Momordica subangulata* and *Naragamia alata*. Indian J pharmacol. 2001;33:276.
9. Choudhury BH, Borah RC, Baruah IC, Baruah NC. Biochemical characterization of traditional medicinal plants used against jaundice caused by Hepatitis virus. Annual Meeting, Society of Biological Chemist 2003.
10. Tesehke CB, Vierke FW, Goldermann L. Carbon tetrachloride levels and serum activities of liver enzymes following acute carbon tetrachloride intoxication. Toxicol Lett. 1983;17:175
11. Kapur V, Pillai KK, Husain SZ, Balbani DK. Hepatoprotective activity of "Jigrine" on liver damage caused by alcohol, carbon tetrachloride and paracetamol in rats. Indian J. Pharmacol. 2000;32:288.
12. Reddy BP, Murthy VN, Venkateshwarlu V, Kokate CK, Rambhau D. Antihepatotoxic activity of *Phyllanthus niruri*, *Tinospors cordifolia* and *Ricinus cumunis*. Indian drugs. 1995;30:7.

13. Borah R C, Choudhury B H, Mohan P. Hepatoprotective activity of *Phyllanthus fraternus* L. And Glycosmis pentaphylla Correa used against Jaundice 2004;In Bioprospecting of Commercially Important Plants 259.
14. Deleve L D, Kaplowitz N. Mechanism of drug-induced liver diseases. Gastroenterol Clin N. Am, 1995; 24: 787-810.
15. Rajesh MG, Latha MS. Heatoprotection by *Elephantopus scaber* Linn. In CCl_4- induced liver injury, Indian J. Physiol Pharmacol. 2001;45(4):481.
16. Kaplowitz N, Aw TY, Simon FR, Stoiz A. Drug Induced hepatotoxicity. Ann Int. Med. 1986;104:826.
17. Ferrel GC. Liver diseases caused by drugs, anesthetics and toxins. In, Sleisenger & Fordtrans Gastrointestinal and liver diseases: Pathophysiology/Diagnosis/ Management. 2nd ed.1998;1221-1253
18. Brent JA, Rumack BH. Role of Free radicals in toxic hepatic injury II. Clin Toxicol. 1993;31:173.