Research Paper Open Access

ARTICLE LINK

ISSN: 2349-9818

SEPTEMBER - OCTOBER 2022, VOL. 9 (5), 45-54

TROPICAL JOURNAL OF PHARMACEUTICAL AND LIFE SCIENCES

(An International Peer Reviewed Journal)

Journal homepage: http://informativejournals.com/journal/index.php/tjpls

HEPATOPROTECTIVE POTENTIAL OF VARIOUS EXTRACTS OF COCCINAINDICA, SIDACORDATA, MEDICAGO SATIVA AND WEDELIATRILOBATA

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ARTICLE INFO:

Received: 9th Sept. 2022**; Received** in revised form: 12th Oct.. 2022**; Accepted:** 19th Oct. 2022**; Available online:** 29th Oct. 2022.

ABSTRACT

Background:

The liver is a crucial organ that is important for the body's removal of xenobiotics. The pharmaceutical business, as well as healthcare practitioners, are challenged by diseases that affect the liver. Botanical medications are frequently employed since the standard therapy for liver problems is linked to a wide variety of negative effects.

Objective - To evaluate the hepatoprotective activity of the various extracts of *Coccinaindica*, *Sidacordata*, *Medicago sativa* and . *Wedeliatrilobata* leaves in mice.

Methods:

The chosen plant parts has been grounded to the fine powder form and this powder is then sieved with a particular number of sieve that is number 40 so as to obtain a uniform powder. Further to this this powder is under gone with WHO guideline.It was found that the specified limits were within all the measured parameters. After physicochemical characterization, all of the plant components were extracted using Pet ether., Chloroform, Ethanol and Aqueous.The evaluation was designed to understand the pharmacological activity at various dose levels of each plant substance, in order to select the appropriate doses.

Results :

Extremely reactive free radical CCl3 specifically targets polyunsaturated endoplasmic reticulum fatty acids in hepatocytes and thus causes over production of Aspartate aminotransferase, Alkaline phosphatase and alanine amino transferase and serum bilirubin.serum enzyme levels of when CCl4 was used to cause liver toxicity increased significantly. of Aspartate aminotransferase, Alkaline phosphatase and alanine amino transferase,Serum bilirubin (T and D) and decrease the levels of total protein. The biochemical evaluation of the liver enzymes showed a potential hepatoprotective effect of Alcoholic (Ethanolic) extract of *Cocciniaindica* (CIEE); Aq. extract of *Sidacordata*(SCAE), Alcoholic (Ethanolic) extract of

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Wedeliatrilobata(WTEE) and ethanolic extract of Medicago sativa(MSEE) in the experimental rat models. The selected medicinal plants are widely employed in various liver disorders in traditional systems of medicine. The present pharmacological evaluation will provide scientific data about the rationale behind their use.

Keywords: Hepatoprotective, *Coccinaindica*, *Sidacordata*, *Medicago sativa*, *Wedeliatrilobata*

INTRODUCTION

Several ways for obtaining drug discovery molecules have been utilized, including plant isolation and various herbal sources. Even if novel chemical buildings are now not developed throughout drug improvement from medicinal plants, current compounds with clean organic recreation may additionally supply essential drug outcomes. The liver is a crucial organ that is important for the body's removal of xenobiotics. The pharmaceutical business, as well as healthcare practitioners, are challenged by diseases that affect the liver. Botanical medications are frequently employed since the standard therapy for liver problems is linked to a wide variety of negative effects. Many of the drugs are available as curing the hepatic disorders in the various texts of the ayurveda. There are various other indigenous plants that are used to treat the liver problems other than those mentioned in the ayurvedic texts. *Coccinaindica*, *Sidacordata*, *Medicago sativa*, *Wedeliatrilobata are* investigated for scientific validation of its folk use in liver disorders.

MATERIAL AND METHOD

Plant Material

Coccinaindica leaf material was collected from Local Vegetable Market, Indore,Madhya Pradesh. *Sidacordata* leaf material was collected from Herbal garden, Government Agriculture College, Mandsaur, Madhya Pradesh. *Medicago sativa* leaf material was collected from Medicinal Plants Garden, Campus of Acropolis Institute of Pharmaceutical Education & Research, Indore. *Wedeliatrilobata* leaf material was collected from Medicinal Plants Garden, Campus of Acropolis Institute of Pharmaceutical Education & Research, Indore

Plant material (leaves) selected for the study were washed thoroughly under running tap water and then rinsed in distilled water; they were allowed to dry for some time at room temperature. Then the plant material was shade dried without contamination for about 3 to 4 weeks.

Experimental Animals

From the pharmacological department of the Acropolis Institute of Pharma. Education and Research, Indore (AIPER, Indore) (Reg. No. 1339/ac/10/CPCSEA) the Adult albino rats weighing 185-225 gms and swiss albino rats weighing 20-35 gms of either sex were used for the experiment.

Preparation of Plant Extract

The powdered plant materials were placed in a 5 liter conical flask with various solvents like $CHCl_3$, Ethanol, Pet.ether and water distilled for imbibing. The Maceration method of extraction was used for the extraction. The vessel is sealed and held for seven days, with some shaking. The liquids were strained after efficient extraction, and marc was pressed, adding the expressed liquid to the strained liquids. Decanting or filtering were used to clarify the mixed liquid. The mixer of solution were concentrated an a water bath and then dried out.

Administration and Dosing of animals

Young albinorats of any sex, balancing 160-200 gms (Wister strain) (10 to 15 weeks old) were used With 3 animals in each group, a total of 16 groups were formed.

Housing and Feeding Condition

The temperature in the testing room was about 25 degreecentigrate. The series was artificially lit with 12 hours of darkness followed by 12 hours of light. Free access to drinking water was fed into the traditional laboratory diet.

Selection of Animals

All the animals selected were labelled for identification and for five days kept in respective cages so that they get acquinted with the laboratory environment.

Preparation of doses

Distilled water was used for preparing all the doses. 1 percent of Tween 80 was used for all extracts. Both concentrations were formulated at 1 ml/100g of body weight in all situations.

Administration of Doses

The dosing was done intraperitoneally that is by intra gastric tube. Animals were kept on fasting before dosing was done for almost 3-4 hours and after that their weight was taken. Post weighing the drug was administered. Again after dosing the food was not given for 3-4 hours.

In each stage 3 animals from each group employed. The research started at a body weight of 300 mg/kg and concluded at a body weight of 5000 mg/kg. In subsequent pharmacological trials, one thenth this dose was measured as the safe. (therapeutic dose). Primarily, animals were monitored at least once every 30 minutes for the first 24 hours following treatment on a daily basis. Death in the whole sample was not identified in any case

Hepatoprotective Activity

To determine the protective effect of liver cells of all extracts of *Cocciniaindica leaves*, leaves of *Sidacordata*, leaves of *Medicago sativa*, leaves of *Wedeliatrilobata* parameter like serum Alanine transferase, Alanine phosphatase, Aspartate amino transferase, Bilirubin in serum (Total and Direct), serum total protein and histopathological studies were carried out. This study was performed for the selection of pharmacologically active extracts. Because CCl4 is readily available in its pure form and consistently causes liver damage in numerous species, including mouse, rat, rabbit, and man, it was employed to induce hepatotoxicity. The type and severity of the liver harm produced can differ from triacylglycerol accumulation, through necrosis to cirrhosis and cancer depending on the dose and mode of application of CCl_4 induced.

As a result of its transformation into a highly reactive poisonous free radical CCl3, the toxic effects of CCl4 are by cytochrome P450, alkylates cellular proteins and is a free radical and in the presence of oxygen, various macromolecules attack polyunsaurated fatty acids at the same time resulting in the lipid peroxides and liver injury. Hepatocellular necrosis causes an increase in serum marker enzymes, which are released from the liver into the blood stream.

RESULT AND DISCUSSION

The findings obtained for the biochemical parameters are comparable with the normal hepatoprotective drug Silymarin, which is as follows;

Control group

Control group was treated with vehicle showed no significant changes in biomarkers enzymes (serum Aspartate aminotransferase, Alkaline phosphatase and alanine amino transferase and serum bilirubin) and total protein levels.

CCl_4 treated group

The group treated with CCl4 reported an improvement in serum levels of Aspartate aminotransferase, Alkaline phosphatase and alanine amino transferase and serum bilirubin. (Total and

Direct). The characteristics feature of experimental hepatic damage observe is significant decrease in protein level.

Silymarin treated group

Group treated with *Silymarin* (Std.) showed highest significant reduction in elevated level of Aspartate aminotransferase, Alkaline phosphatase and alanine amino transferase, bilirubin (T and D) and a highest significant increase in total protein level. Percentage protection (H) produced by the *Silymarin* on the reduction of Alanine amino transferase, total bilirubin, aspartate aminotransferase, and phosphatase alkaline and Direct bilirubin levels were 82.48%, 96.98%, 88.19%, 86.85% and 86.16% respectively. A substantial rise was 76.11 percent in the overall level of protein.

Cocciniaindica

Group treated with Ethanol extract (CIEE) showed significant reduction in elevated level of Aspartate aminotransferase, Alkaline phosphatase and alanine amino transferase and bilirubin. Percentage protection produced by the CIEE on the reduction of 39Alanine amino transferase, total bilirubin, aspartate aminotransferase, and phosphatase alkaline and Direct bilirubin levels were 67.06%, 74.81%, 78.34%, 76.32% and 76.93% respectively. 64.68 percent was a substantial improvement in the overall amount of protein .

Group treated with petroleum ether extract (CIPE) shown moderate decrease in enzyme levels. Percentage protection produced by the CIPE on the reduction of 39Alanine amino transferase, total bilirubin, aspartate aminotransferase, and phosphatase alkaline and Direct bilirubin levels were 29.10%, 40.94%, 57.20%, 52.64% and 61.54% respectively.A significant increase in total protein level was 59.74%.

Chloroform extract (CICE) and Aqueous extract (CIAE) however didn't show any reduction in enzyme level significantly, indicating no activity.

Sidacordata

Group treated with Aqueous extract (SCAE) showed significant reduction in elevated level of Aspartate aminotransferase, Alkaline phosphatase and alanine amino transferase and bilirubin. Percentage protection (H) produced by the SCAE on the reduction 39Alanine amino transferase, total bilirubin, aspartate aminotransferase, and phosphatase alkaline and Direct bilirubin levels were 62.20%, 65.56%, 70.88%, 67.55% and 72.31% respectively.A important 63.12 percent rise in the overall level of protein was found. Group treated with Ethanol extract (SCEE) shown moderate decrease in enzyme levels. Percentage protection produced by the SCEE on the reduction 39Alanine amino transferase, total bilirubin, aspartate aminotransferase, and phosphatase alkaline and Direct bilirubin levels were 30.26%, 30.42%, 39.23%, 55.27% and 52.31% respectively.A significant increase in total protein level was 46.50% Chloroform extract (SCCE) and Pet ether extract (SCPE) however didn't show any reduction in enzyme level significantly, indicating no activity.

Medicago sativa

Group treated with Ethanol extract (MSEE) showed significant reduction in elevated level of Aspartate aminotransferase, Alkaline phosphatase and alanine amino transferase and bilirubin. Percentage protection (H) produced by the MSEE on the reduction of 39Alanine amino transferase, total bilirubin, aspartate aminotransferase, and phosphatase alkalineand Direct bilirubin levels were 64.66%, 85.20%, 77.37%, 75.44% and 72.30% respectively. A significant increase in total protein level was 61.30%. Group treated with Aqueous extract (MSAE) shown moderate decrease in enzyme levels. Percentage protection produced by the MSAE on the reduction 39Alanine amino transferase, total bilirubin, aspartate aminotransferase, and phosphatase alkaline and Direct bilirubin levels were 33.08%, 64.35%, 54.01%, 64.92% and 52.31% respectively.A significant increase in total protein level was 44.42%

Group treated with Chloroform extract (MSCE) also shown moderate decrease in enzyme levels. Percentage protection (H) produced by the MSCE on the reduction of 39Alanine amino transferase, total bilirubin, aspartate aminotransferase, and phosphatase alkaline and Direct bilirubin levels were 25.36%, 38.80%, 31.46%, 43.86% and 64.61% respectively.A significant increase in total protein level was 28.58%

Pet ether extract (MSPE) however didn't show any reduction in enzyme level significantly, indicating no activity.

Wedeliatrilobata

Group treated with Ethanol extract (CIEE) showed significant reduction in elevated level of Bilirubin, alanine amino transferase, aspartate aminotransferase, and alkaline phosphatase. Percentage of reduction in aspartate aminotransferase, alkaline phosphatase, alanine amino transferase, and total bilirubin produced by the WTEE and Direct bilirubin levels were 67.06%, 74.81%, 78.34%, 76.32% and 76.93% respectively. 64.68 percent was a substantial improvement in the overall amount of protein .

Group treated with petroleum ether extract (CIPE) shown moderate decrease in enzyme levels. Percentage protection produced by the WTPE on the reduction of direct level of "bilirubin, total bilirubin, Aspartate aminotransferase, Alkaline phosphatase and alanine amino transferase" levels were 29.10%, 40.94%, 57.20%, 52.64% and 61.54% respectively.A significant increase in total protein level was 59.74%.

Chloroform extract (CICE) and Aqueous extract (CIAE) however didn't show any reduction in enzyme level significantly, indicating no activity.

Comparative AST Level for Different Extracts, Silymarin (Std.) and Positive Control Group in CCl4 provoked Hepatotoxicity in Rats

S. No.	Treatment	Mean Serum AST level (IU/L) ± SEM			
		Pet. Ether	Chloroform	Ethanol	Aqueous
1.	*Cocciniaindica*	788.42 ±12.18#	892.74 ±11.90	558.24 ±8.34#	908.68 ±8.94
2.	*Sidacordata*	960.54 ±13.80	924.74 ±12.12	781.34 ±13.64#	587.68 ±11.69#
3.	*Medicago sativa*	947.50 ±14.32	811.39 ±9.37#	572.80 ±11.32#	764.23 ±9.57#
4.	*Wedeliatrilobata*	748.32 ±9.08#	932.82 ±6.02	575.9 ±8.64#	961.54 ±10.10
4.	Control	358.50±8.96			
5.	CCl_4 treated	964.74±12.64*			
6.	Silymarin [Std.]	465.680±9.26#			

Each group`s value are the meanS.E.M. Of six animals.

$P < 0.05$ was considered as significant.

* $P < 0.05$ when compared with Control group.

# $P < 0.05$ when compared with CCl_4 treated group.

Of six animals, 30E.M. Significant was defined as P 0.05.

When compared to the Control group, 42P 0.05. # P 0.05 when compared to the group that received CCl4.

Comparative ALT Level for Different Extracts, Silymarin (Std.) and positve Control Group in CCl4 provoked Hepatotoxicity in Rats

S. No.	Treatment	Mean Serum ALT level (IU/L) ± SEM			
		Pet. Ether	Chloroform	Ethanol	Aqueous
1	*Cocciniaindica*	428.33 ±8.67#	602.55 ±9.43	267.55 ±8.27#	577.67 ±9.36
2	*Sidacordata*	576.24 ±9.53	594.40 ±6.84	478.13 ±11.12#	311.42 ±8.43#
3	*Medicago sativa*	580.37 ±14.18	438.38 ±13.76#	218.24 ±7.81#	317.16 ±12.76#
4	*Wedeliatrilobata*	486.23 ±10.2#	598.62 ±8.43	303.7 ±6.43#	487.70 ±9.56
4	Control	148±5.57			
5	CCl_4 treated	622.43±12.58*			
6	Silymarin [Std.]	162.33±14.9#			

Each group`s value are the mean S.E.M. Of six animals

$P < 0.05$ was considered as significant.

* $P < 0.05$ when compared with Control group.

# $P < 0.05$ when compared with CCl_4 treated group.

P 0.05 was deemed to be significant for the 30mean S.E.M. of the six animals.

When compared to the Control group, 42P 0.05. # P 0.05 when compared to the group that received CCl4.

Comparative ALP Level for Different Extracts, Silymarin (Std.) and Positive Control Group in CCl4 provoked Hepatotoxicity in Rats

S. No.	Treatment	Mean Serum ALP level (IU/L) ± SEM			
		Pet. Ether	Chloroform	Ethanol	Aqueous
1	*Cocciniaindica*	527.35 ±7.99#	668.36 ±10.28	448.71 ±8.76#	710.5 ±11.88
2	*Sidacordata*	680.53 ±8.37	715.32 ±8.67	594.22 ±11.12#	476.45 ±7.54#
3	*Medicago sativa*	727.38 ±11.4	623.13 ±7.29#	452.31 ±8.78#	539.23 ±9.62#
4	*Wedeliatrilobata*	560.35 ±6.68#	626.28 ±5.68	468.62 ±7.62#	690.6 ±10.68
4	Control	368.11±7.43			
5	CCl_4 treated	740.16±37.15*			
6	Silymarin [Std]	412.06±6.79#			

Each group`s value are the mean S.E.M. Of six animals

$P < 0.05$ was considered as significant.

* $P < 0.05$ when compared with positive Control group.

# $P < 0.05$ when compared with CCl_4 treated group.

Comparative Serum Bilirubin (Total) Level for Different Extracts and Control Group in CCl4 induced Hepatotoxicity in Rats

S. No.	Treatment	Mean Serum Bilirubin (T) level (mg/dL) ± SEM			
		Pet. Ether	Chloroform	Ethanol	Aqueous
1	*Cocciniaindica*	1.154 ±0.032#	1.623 ±0.045	0.886 ±0.056#	1.694 ±0.054
2	*Sidacordata*	1.694 ±0.029	1.621 ±0.038	1.120 ±0.022#	0.980 ±0.029#
3	*Medicago sativa*	1.657 ±0.087	1.250 ±0.027#	0.893 ±0.036#	1.019 ±0.067#
4.	*Wedeliatrilobata*	1.201 ±0.061#	1.512 ±0.034	0.764 ±0.045#	1.582 ±0.043
4	Control	0.617±0.021			
5	CCl_4 treated	1.761±0.047*			
6	Silymarin [Std]	0.768±0.027#			

Each group`s value are the mean S.E.M. Of six animals.
$P < 0.05$ was considered as significant.
* $P < 0.05$ when compared with positive Control group.
# $P < 0.05$ when compared with CCl_4 treated group.

Comparative Serum Bilirubin (Direct) Level for Different Extracts and Control Group in CCl4 induced Hepatotoxicity in Rats

S. No.	Treatment	Mean Serum Bilirubin (D) level (mg/dL) ± SEM			
		Pet. Ether	Chloroform	Ethanol	Aqueous
1	*Cocciniaindica*	0.486 ±0.018#	0.798 ±0.037	0.386 ±0.017#	0.832 ±0.029
2	*Sidacordata*	0.853 ±0.015	0.813 ±0.032	0.542 ±0.18#	0.417 ±0.014#
3	*Medicago sativa*	0.846 ±0.024	0.658 ±0.016#	0.413 ±0.036#	0.542 ±0.019#
4.	*Wedeliatrilobata*	0.375 ±0.021#	0.687 ±0.026	0.275 ±0.016#	0.721 ±0.018
4	Control	0.234±0.019			
5	CCl_4 treated	0.887±0.039*			
6	Silymarin [Std]	0.326±0.012#			

Each group`s value are the mean S.E.M. Of six animals
$P < 0.05$ was considered as significant.
* $P < 0.05$ when compared with positive Control group.
# $P < 0.05$ when compared with CCl_4 treated group.
thirty mean SEM of six animals When compared to the positive Control group, P 0.05 was regarded as significant. # P 0.05 when compared to the group that received CCl4.

Comparative Serum Total Protein (TP) Level for Different Extracts and Control Group in CCl4 induced Hepatotoxicity in Rats

S. No.	Treatment	Mean Serum Total Protein level (g/dL) ± SEM			
		Pet. Ether	Chloroform	Ethanol	Aqueous
1	*Cocciniaindica*	5.374 ±0.023#	4.014 ±0.019	6.319 ±0.034#	3.918 ±0.025
2	*Sidacordata*	4.012 ±0.029	4.298 ±0.036	5.617 ±0.028#	6.258 ±0.035#
3	*Medicago sativa*	3.973 ±0.029	4.925 ±0.022#	6.182 ±0.039#	5.538 ±0.031#
4.	*Wedeliatrilobata*	4.262 ±0.012#	3.012 ±0.018	7.218 ±0.022#	2.818 ±0.012
4	Control	7.674±0.021			
5	CCl_4 treated	3.821±0.044*			
6	Silymarin [Std]	6.751±0.036#			

Each group`s value are the mean

Of six animals, 30E.M. Significant was defined as P 0.05.

$P < 0.05$ was considered as significant.

* $P < 0.05$ as compared to positive Control group.

# P < 0.05 when compared to the CCl4 treated group.

CONCLUSION

The source of rakthavaha and ranjakapitta is the liver (Vakrit) in Ayurveda. As per ayurveda the main factor for the development of any disease in the body is damage to the liver which inturn damage the digestion. Many of the drugs are available as curing the hepatic disorders in the various texts of the ayurveda. There are various other indigenous plants that are used to treat the liver problems other than those mentioned in the ayurvedic texts.

The present study is an exhaustive summary of our findings on the inherent potential of indigenous plants are used as medicine to treat for treating liver toxicity with a full shield and effectiveness profile in animals. The hepatoprotective effect of the plants is likely because of the presence of flavonoids, alkaloids, terpenoids, glycosides and steroids. Result obtained from hepatoprotective activity screening indicate that Ethanolic extract of *Cocciniaindica* (CIEE); Aqueous extract of *Sidacordata*(SCAE), ethanolic extract of Medicago sativa (MSEE) and Ethanolic extract of *Wedeliatrilobata*(WTEE) shows more prominent results as hepatoprotective.

ACKNOLEDGEMENT

The authors are would like to acknowledge the helpand support stretched by the Department of Pharmacy, Acropolis Institute of pharmaceutical Education and Research in Indore MP.

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How to cite this article: Mantry Piyush, and Dr. Jat K. Rakesh. "HEPATOPROTECTIVE POTENTIAL OF VARIOUS EXTRACTS OF COCCINAINDICA, SIDACORDATA, MEDICAGO SATIVA AND WEDELIATRILOBATA". *Tropical Journal of Pharmaceutical and Life Sciences*, vol. 9, no. 5, Oct. 2022, p. 45-54, https://informativejournals.com/journal/index.php/tjpls/article/view/110.

PUBLISHED BY:
INFORMATIVE JOURNALS
JADOUN SCIENCE PUBLISHING GROUP INDIA