International Journal for Multidisciplinary Research (IJFMR)



E-1

E-ISSN: 2582-2160 • Website: <u>www.ijfmr.com</u>

• Email: editor@ijfmr.com

To Evaluate the Hepatoprotective Activity of Leaves of Salacia Chinensis Linn on CCl₄ Induced Albino Wistar Rats

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Abstract:

The main aim of the present study is to evaluate the Hepatoprotective activity of aqueous slurry of leaves of *Salacia chinensis* Linn .on CCl₄ induced liver damage in rats. Hepatotoxicity was induced by CCl₄ and the biochemical parameters such as serum glutamate pyruvate transminase (sGPT), serum glutamate oxaloacetate transaminase (sGOT), alkaline phosphatase (sALP), Blood glucose level, Liver glycogen level and histopathological changes in liver were studied along with silimarin as standard Hepatoprotective agents. The Phytochemical investigation of the aqueous slurry showed significant decrease in the levels of sGPT, sGOT, sALP as compared to the animals treated with Silimarin, a known, standard hepatoprotective agent. A significant decrease in blood sugar level and liver glycogen was also observed in the animals treated with aqueous slurry of leaves of *Salacia chinensis* Linn.

Keywords: CCl₄, Hepatoprotective, sGPT, sGOT, Salacia chinensis Linn.

Introduction

In India and in South East Asia, incidence of liver dysfunction is more than in any developed countries, and hence there is a greater need for a cure for liver disorders such as Jaundice and Hepatitis. Today the world over, there is a great deal of interest in Ayurvedic system of medicine and thus there is an ever increasing demand for various commonly used medicinal plants for the production of ayurvedic medicines (IDMA Bulletin 1998).

Liver disorders:-

Liver is a key organ in regulating homeostasis of the body. The diversity of physiological roles means that, if liver function is impaired, several homeostatic metabolisms will be affected. Some of the liver diseases are Acute liver disease, causing fluminant hepatic failure, chronic liver disease, leading to liver cirrhosis and Acute hepatitis.

Hepatoprotection can be evaluated by studying various marker enzymes (Zimmerman 1978) Determination of activity of hepatic enzymes released into the body by damaged liver is one of the most useful tools in the study of hepatotoxicity (Zimmerman 1978). These enzymes are generally called liver marker enzymes. These are alkaline phosphates, Aspartate Amino Transferase (AST), Alanine Amino Transferase (AST) (Zimmerman 1978).

Many plants have been used for the treatment of liver disorders in folk remedies. A few of these are stem bark of *Azadirecta indica*, *Tinospora cordifolia*, *Picorrhiza kurroa*, *Silybum marianum*, *Phyllanthus*



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species, *Swertia chiraita* and *Ricinus communis*. (Patel, Saluja 2000). The roots of *Salacia chinensis* commonly known as Saptarangi, belonging to the family Hippocrataceae also show medicinal properties (Yadav and Sardesai 2002). The roots of *Salacia chinensis* have biological active compounds and colouring agents which show various activities like antioxidant and skin lightening agent. (Deokate and Khadbadi 2012). Ethanolic extract of roots of *Salacia chinensis* is found to be hepatoprotective (Agusti 2010).

The present work will help us to evaluate the hepatoprotective properties of leaves of *Salacia chinensis*, and it will be compared with Silymarine, a known, well documented hepatoprotective monoherbal formulation. The study was carried out on CCl₄ induced albino Wister rats.

Materials and Methods:-

Healthy matured leaves of *Salacia chinensis* were washed thoroughly to remove dust and other extraneous matter. The excess of water is absorbed by spreading the plant material over filter paper for three days in shade, away from sunlight. The filter paper was replaced daily. The leaves that were turned yellow were discarded, the rest of the leaves were then placed in a preset oven and incubated at $45^0 \pm 5^{\circ}$ C. Fully dried green leaves were powdered, sieved through BSS mesh 85 and then stored in air tight purl pet transparent jars, the jars were labeled properly, this leaf powder was used in the present investigations, for hepatoprotective activity using albino rats (250-300gms) of either sex.

Procurement and maintenance of animals:-

Animals used for efficacy study were Albino Wister rats. Animals (female rats), after procurement were segregated, weight wise, in polyuretane cages. In a cage three animals were housed. The cage was provided with rice husk bedding. The animals were provided with drinking water *ad libitum* and were fed on commercially available mice feed.

Dosage:-

Reversible liver damage was induced by an intra-peritoneal (i.p.) injection of CCl₄, at a dose of 0.7ml/kg (Pandey and Chaturvedi.1969) in 0.5ml liquid paraffin as a vehicle to animal from Group **II** to V. An i.p. injection of 0.5ml/kg liquid paraffin was given to each from Group I.

An oral dose of 500mg/kg of leaf powder of *Salacia chinensis* was administered to each animal of Group IV.

A dose of 0.07g/kg silymarin (in the form of Silybon tablets) suspended in 2 ml distilled water was administered to each rat of Group V. The normal control, CCl₄ control and CCl₄ recovery group animals were also administered 2 ml of distilled water each as sham treatment.

The mode of dosing was oral via a gavage. The animals were first given CCl₄ injection intraperitonially, then the oral dose of leaf powder of leaf powder *Salacia chinensis* and silymarin group.

Animals from Group I, II IV and V were sacrificed at 72 hours after CCl₄ damage [(period of maximum damage) Munro and Fleck, 1967. Zimmerman, 1978. Timbell 1982. Poole and Leslie 1989)]. The animals from Group III were sacrificed on seventh day of the study.



Dose regimen

Days	Group I	Group II	Group III	Group IV	GroupV CCl ₄ +
	Normal Control	CCl ₄ Control	CCl ₄ Recovery	CCl ₄ +Leaf powder	Silymarin
1	0.5ml liq paraffin	0.7ml/Kg	0.7ml/Kg CCl ₄	0.7ml/Kg CCl ₄ in	0.7ml/Kg CCl ₄
	ip+ 2ml DW OD	CCl_4 in 0.5	in 0.5 ml	0.5ml	in0.5ml
		ml	Liq.Paraffine	Liq.Paraffine ip &	Liq.Paraffine ip
		Liq.Paraffine	ip+2ml DW	500mg/kg of leaf	& 0.007g/Kg
		ip+2ml DW	OD	powder in 2ml	Silymarin in
		OD		DW OD	2ml DW OD
2	2 ml DW OD	2 ml DW OD	2 ml DW OD	500mg/kg of leaf	0.007g/Kg
				powder in 2ml	Silymarin in
				DW OD	2ml DW OD
3	2 ml DW OD	2 ml DW OD	2 ml DW OD	500mg/kg of leaf	0.007g/Kg
				powder in 2ml	Silymarin in
				DW OD	2ml DW OD
4	Sacrifice	Sacrifice	2 ml DW OD	Sacrifice	Sacrifice
5			2 ml DW OD		
6			2 ml DW OD		
7			Sacrifice		

Parameters observed:

Cage side observations included general behavioral changes, daily food and water consumption and daily weight changes. Along with blood and tissue (Liver) biochemistry parameters, histopathological evaluations of the liver under light microscopy were also done.

Sacrifice: Food was discontinued 18 hours prior to the sacrifice for each group but water was provided *ad libitum*. Blood was withdrawn by using butterfly needle no 16 Weights of the animals were recorded before sacrifice.

Blood samples were analyzed (within 8 hours of collection) for blood biochemical parameters, like levels of SGPT, SGOT and AlkPO₄. Histopathological evaluations of the liver under light microscopy were also done.

	SGOT	SGPT	Alk	Bilirubin
			PO_4	
Grp I	7.73	2.10	1.45	0.43
Grp II	19.27	25.56	5.04	0.48
Grp	11.97	18.22	3.12	0.46
III				
Grp	8.54	11.71	2.06	0.45
IV				
Grp	10.56	14.92	14.52	0.45
V				

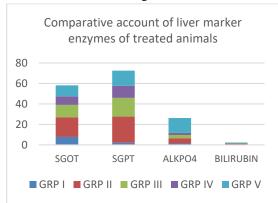
Observations: Table 01 Average values of liver	marker enzymes
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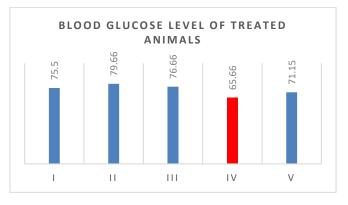
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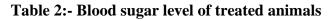




Biochemical parameters: - (Table 1 and Graph 1)

CCl₄ treatment caused significant increase in plasma SGOT and SGPT levels. These levels were not recovered significantly after the natural recovery phase. Treatment with aqueous slurry of leaf powder of Salacia chinensis caused significant reduction in SGOT, SGPT levels. These levels were significantly high in the animals of CCl₄ control group (Grp II), indicating the toxic effect of CCl₄. Level of Alkaline Phosphatase in the animals treated with CCl₄ (Grp. II) showed a significant increase as compared to the normal control group (Grp I). However the animals treated with the aqueous slurry of Salacia chinensis showed a significant decrease in the level of Alkaline Phosphatase, which is much low than that of the animals treated with Silibon (Grp V).





	Sugar
Grp	level
Ι	75.5
II	79.66
III	76.66
IV	65.66
V	71.15

Blood sugar level of the treated animals showed a significant decrease in group IV (animals treated with aqueous extract of leaf powder of *Salacia chinensis* as compared to group II (CCl_4 control) and group V (CCl_4 + Silimarin)



Discussion:

There were no significant changes in the behavior of any of the treated animals from all the treatment groups. Food and the water consumption decreased after CCl₄ treatment. This is an indication of toxic response by the treated animals. Decrease in the body weight after CCl₄ treatment in treated animals, further confirmed the toxic response. Increase in the body weight, after the treatment of aqueous slurry of leaf powder of *Salacia chinensis*; indicate the protective action of the treatment, to CCl₄ induced damage. It is reported that peak changes in plasma enzymes are noticed at 24 hours after CCl₄ administration, but complete recovery is slow and takes about 14 days (Zimmerman. 1978).

Alternation of serum enzyme levels can be monitored to evaluate the hepatocellular damage caused by various foreign compounds (Grice et, al. 1971. Molander and Sheppard, 1957. Worbelwoski and LaDue, 1956). CCl₄ treatment caused a significant increase in blood and tissue parameters studied. CCl₄ caused a marked elevation in the transaminases (Subbarao and Gupta, 1971).

Conclusion:

Aqueous extract of leaf powder of *Salacia chinensis* Lin. has shown significant (p<0.05) hepatoprotective activity in CCl₄ induced hepatotoxicity as compared to the treatment with Silimarin. Aqueous extract of leaves of *Salacia chinensis* was found to be more effective than the treatment of Silibon. Histopathological studies also confirmed the above investigation

It is interesting to note a significant reduction in blood sugar level and liver glycogen, in the animals treated with aqueous slurry of leaf powder of *Salacia chinensis*. However, more work is needed to be carried out to evaluate glycogen reducing effect of leaf powder of *Salacia chinensis*, using various organic solvents. Plans produce leaves and the phytoconstituents present in the leaves continuously. It is one of the best examples of Bio factories.

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