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A Study on Hepatoprotective Activity of *Pergularia Daemia*.

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ABSTRACT

Liver disease is one of the major problems in developing countries like India. As India is the land of flora, numerous plants are available plenty with medicinal values and they could be used to treat diseases. The present study is aimed to study the efficacy and hepatoprotective activity of *Pergularia daemia* in experimentally induced hepatotoxic albino rats. Twelve animals were chosen for the present study. They were divided into 3 groups as normal, untreated and treated. Each group consists of 4 animals. The overnight fasted rats were injected with 0.3 ml of CCl₄ and with paraffin for 10 days per animal. The crude powder of *Pergularia daemia* was administered orally on the day of hepatotoxic substance CCl₄ injection to the third group (treated) of animals for 10 days. The blood sample and liver homogenate were collected and the liver functions of the 3 group of animals were estimated. The serum and tissue markers were elevated in the untreated group compared to normal group and also the treated group showed the normal levels of enzyme markers as in the normal control group. It is found that *Pergularia daemia* is having hepatoprotective activity.

Keywords: Hepatoprotective, *Pergularia daemia*, CCl₄.

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INTRODUCTION

India is a land of medicinal plants. In ancient days, plants were used to prevent and cure diseases. The compounds present in plants have the ability to reduce the toxicity of any chemical or a pathogen that enters the human body [1]. Liver is the largest glandular organ in human body. It plays a major role in purifying the blood by eliminating the toxic substances. Liver is the only human organ which is capable of regenerating its lost tissue. There is no mechanism for replacing the function of liver [2]. The chemical substances that affect the function of liver are alcohol and carbon tetrachloride (CCl₄) [3-6]. *Pergularia daemia* commonly called as Trellis vine is a perennial twining herb that belongs to milky weed family Asclepiadaceae [7]. The plant was used as laxative, anti-pyretic, expectorants and also in infantile diarrhea [8]. The latex of the leaf is used as pain killer [8]. The phytochemicals present in the leaves of *Pergularia daemia* are flavanoids, alkaloids, terpenoids, steroids and tannins [9]. Pharmacological studies have confirmed that *Pergularia daemia* exhibit a broad range of biological effects. The crude extract of plant has been used as a traditional medicine for the treatment of various diseases [7]. Hence, the present work is focused towards the study on the efficacy and hepatoprotective activity of crude powder of whole plant to treat the experimentally induced jaundice affected albino rats.

MATERIALS AND METHODS

Healthy male albino rats weighing 150-200 g were purchased from a private agency in Bengaluru and were used for all experiments. The animal experiments were conducted in the Department of Siddha Medicine, Tamil University after the approval of animal ethics committee. They were given commercial diet and tap water *ad libitum*. The diet was purchased from Hindustan Lever Ltd., Bengaluru. The overnight fasted rats were injected with 0.3ml of CCl₄ mixed with paraffin in the ratio of 3:1 for ten days per animal daily. The crude powder was orally administered to hepatotoxic rats. 0.9 percent saline was used as a vehicle and administered to controls. The animals were divided into 3 groups consisting of 4 animals each. The first group was considered as normal and the second as untreated. The third group was administered orally with crude powder of *Pergularia daemia* at a dose of 100 mg/100g body weight. The crude powder administration was initiated on the day of injection of CCl₄ and both were continued up to 10th day. 12 hours before sacrifice, the food alone was withdrawn. On the 11th day the animals were anesthetized and then sacrificed. The blood was collected in dry, well cleaned test tubes and they were kept in refrigerator overnight and clear serum was separated after centrifugation. The liver was perfused with 0.9 percent saline, excised and then homogenized with ice cold saline in a Teflon homogenizer. The total bilirubin, alkaline phosphatase (ALP), acid phosphatase (ACP), lipid peroxide (LPO) in tissue and serum, gamma glutamyl transferase (γ -GT), aspartate transaminase (AST) and alanine transaminase (ALT) in serum were estimated.

RESULTS

In the present experimental study, the normal animals showed 0.26 ± 0.028 mg/dl, as the normal level of serum bilirubin. After the induction of CCl₄ (30 mg/kg) in the rats, it was found to be increased to 0.590 ± 0.052 mg/dl than the normal level. After the treatment with *Pergularia daemia* crude powder at the dose of 100 mg/100 g of body weight, the serum bilirubin level was decreased to 0.200 ± 0.025 mg/dl. (Table) When CCl₄ was injected to the animals, the serum lipid peroxidation and tissue lipid peroxidation levels were elevated to 3.95 ± 0.35 n moles MDA/mg and 5.25 ± 0.42 n moles MDA/mg than the normal level of about 1.60 ± 0.13 n moles MDA/mg and 2.36 ± 0.25 n moles MDA/mg respectively. After the administration of crude powder, the serum lipid peroxidation level and the tissue lipid peroxidation levels were reduced to 1.97 ± 0.21 n moles MDA/mg and 3.40 ± 0.30 n moles MDA/mg which was found to be closer to the normal level of serum lipid and tissue lipid peroxidation levels respectively.

The serum gamma glutamyl transferase level was elevated to 20.00 ± 2.18 U/l when the hepatotoxic substance was injected. The normal serum γ -GT level was found to be 1.00 ± 0.10 U/l and after the treatment with the crude powder of *Pergularia daemia* the level was significantly reduced to 3.00 ± 0.27 U/l.

The normal level of serum alanine transaminase is 90.87 ± 9.63 U/l. After the injection of CCl₄ the serum ALT level was rapidly raised to 197.71 ± 21.55 U/l and it was reduced to the level of 120.40 ± 11.35 U/l by treating the animals with the crude powder of *Pergularia daemia*.

The level of serum aspartate transaminase is 102.84 ± 11.20 in normal animals. When the hepatotoxic CCl_4 was injected the AST level was raised to 294.28 ± 30.01 U/l in the untreated animals and the treated animals showed a significant reduction in their AST levels to about 160.22 ± 12.20 U/l.

The normal level of serum alkaline phosphatase was found to be 81.25 ± 4.79 U/l and this level was raised to 92.20 ± 4.70 U/l when CCl_4 was injected. When the animals were treated with *Pergularia daemia* powder the reduction of serum ALP was to 70.82 ± 4.31 U/l which was found to be nearer to the normal level of ALP.

The normal animals show 87.26 ± 8.90 U/l as the normal level of serum acid phosphatase. After the induction of hepatotoxicity, the level was increased to 99.89 ± 10.68 U/l and the significant reduction in the serum ACP level was observed to about 55.00 ± 0.50 U/l after the treatment with the crude powder of *Pergularia daemia*.

Table: Clinical parameters of various animal groups

Group	Dose	Bilirubin mg/dl	LPO in serum nM/ml MDA	LPO in tissue nM MDA/mg protein	γ -GT U/l	ALT U/l	AST U/l	ALP U/L	ACP U/l
Normal	Isosaline	0.26 ± 0.028	1.60 ± 0.13	2.36 ± 0.25	1.00 ± 0.10	90.87 ± 9.63	102.84 ± 11.20	81.25 ± 4.79	87.26 ± 8.9
CCl_4 treated	0.3 ml	0.590 ± 0.052	3.95 ± 0.35	5.25 ± 0.42	20 ± 2.18	197.71 ± 21.55	294.28 ± 30.01	92.20 ± 4.70	99.89 ± 10.68
Crude powder	100mg/kg	0.200 ± 0.025	1.97 ± 0.21	3.40 ± 0.30	3.00 ± 0.27	121.40 ± 11.35	160.22 ± 12.20	70.82 ± 4.31	55 ± 5
Percentage of changes		66 %	50.13 %	35 %	85 %	38.5 %	45.5 %	23.18 %	44.9 %

DISCUSSION

Liver disease is the devastating disease throughout the world especially in India. The injection of CCl_4 to the healthy rats caused some effects like damage to the liver membrane or liver tissues, loss of structural integrity in the liver cells and dysfunction of hepatic cells [10]. Liver diseases arise due to various other reasons as well. There are plenty of synthetic molecules available to treat liver diseases. However, these synthetic molecules cause adverse events. Moreover, many alcoholic compounds like carbon tetrachloride (CCl_4) have the ability to cause liver diseases. In CCl_4 , the active metabolite namely trichloromethyl radical induces the lipid peroxidative degradation of the membranes of liver [11]. This influences the increase of serum markers like total bilirubin, ALT, AST and ALP. Among these serum markers, ALT, AST and ACP are widely used in the detection of damage to the liver. The AST and ALT diffuses out of the liver, when the liver membrane gets damaged and the ACP diffuses out of the liver when the tissue gets damaged [12]. Thus, these serum markers get distributed throughout the body and are found in increased levels in the serum. Hence these are better markers to detect the liver injury. The ALP serum marker is associated with the hepatic cell function. If there is an increase in biliary pressure, then there will be an increase in the ALP level [13]. The intra hepatic biliary obstruction leads to the increase in γ -GT levels in serum. The increase in LPO is due to the loss of structural integrity in hepatocytes [14]. The present study showed that the oral administration of the crude extract of *Pergularia daemia* helps to treat and protect the hepatic cells and resulted in the decreased levels of these serum markers which were statistically significant. The similar results were reported by Jain.S.C et.al. [15]. In the present study, the obtained results are assumed to be due to the presence of flavanoids in the crude extract of the plant and it was reported in the earlier study [16]. We have studied the effect of CCl_4 on the liver of rats and the treatment of crude extract of *Pergularia daemia* on the affected rats. This study can be done on other organisms and the effect of the crude extract on those organisms can also be studied. In conclusion, the oral administration of crude powder of *Pergularia daemia* has more efficacy towards hepatoprotective activity. Further studies are needed to confirm our results.

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