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# Investigation of Hepatoprotective Activity of *Passiflora nepalensis*

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**Abstract:** This study was intended to evaluate the hepatoprotective activity of the whole plant of *Passiflora nepalensis*. The hepatoprotective activity study was carried out by using simvastatin (20mg/kg p.o) induced hepatotoxicity. The methanolic extract of whole plant of *Passiflora nepalensis*was administered at different doses such as 150 and 225 mg/kg body weight and the study was compared with standard drug Silymarin (25mg/kg). The extract exhibited significant hepatoprotective activity, which supports the traditional medicinal utilization of the plant.

**Key words:** *Passiflora nepalensis* · Hepatoprotective · Simvastatin · Edema

## INTRODUCTION

There are 500 species in the Genus Passiflora. In 1529 Spanish discovered Passiflora which comes from a Latin word "Passio" which was a symbol for "Passion of Christ [1]. They are common for warm and tropical areas [2]. The synonyms of *Passiflora nepalensis* are Passion fruit (English), Krishna Fal (Hindi), Pansara (Bengali) and Garindal (Nepali) [3]. In Eastern India, *Passiflora nepalensis* is commonly found which belongs to the family Passifloraceae [1]. It is commonly found in Sikkim, Himalayan region, Assam and Bengal of India [3]. Various chemical constituents are present in Passiflora like phenols, glycosyl flavonoids and cyanogenic compounds. Vitexin has been isolated from methanolic extract of *Passiflora nepalensis* which is one of the crucial glycosyl flavonoid [1- 3]. Isovitexin, orientin, isoorientin, apigenin, kaempferol and quercetin, carbohydrates, amino acids, benzopyrones, cyanogenic glycosides such as gyanocardin, pyrone derivatives such as maltol and ethyl maltol are also the chemical ingredients of Passion flower [4]. The medicinal value of Passion flower is present in Ayurveda, Siddha and Unani systems of medicine [2]. *P. nepalensis* is used as a cure for hypertension and inflammation. It has potent antioxidant activity [5]. *Passifloraedulis* is useful as a sedative and to cure central disorders such as anxiety and insomnia [3]. It has negative chronotropic effects. It has hypolipidemic activity which is stated by rural community of Sikkim State. It also acts as protective agent against renal ischemia/reperfusion [2]. *Passiflora nepalensis* treats anxiety, opiates withdrawal, insomnia, attention-deficit hyperactivity disorder and cancer [4]. The juice of *P. nepalensis* is used in the manufacturing of candy, squashes, cordials, syrups, carbonated beverages and jellies[3]. *Passiflora nepalensis* is grown as an ornamental plant in gardens [5].

The liver regulates homeostasis in the body. Almost all the biochemical pathways related to growth, nutrient supply, energy provision and reproduction takes place in liver. Jaundice and hepatitis are two major hepatic disorders that are responsible for high mortality. Currently only a few hepatoprotective drugs are available for the cure of liver diseases[6]. Treatment options for common liver diseases such as cirrhosis, fatty liver and chronic hepatitis are problematic. Physician and patients are in need of effective therapeutic agents with low tendency of side effects [7]. Herbal drugs play a role in the management of various liver disorders most of which speed up the natural healing processes of the liver.Many herbal plants and their formulations are used for the treatment of liver disorders in traditional system of medicine in India [8].

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Silymarin is a flavonolignan that has been recently introduced as a hepatoprotectiveagent. Silymarin is extracted from the seeds anda fruit of *Silybummarianum* and actually is amixture of three components: Silibinin, Silydianine and Silychristine. Clinical trials have reported that Silymarin shows efficacy in acute viral hepatitis,poisoning by *Amanita phalloides,* ethanol,paracetamol and carbon tetrachloride [8].

## MATERIAL AND METHODS

**Plant Material:** The whole plant of *Passiflora nepalensis* Walp., Passifloraceae, was collected in the month of October from Eastern India (Sikkim Himalayas) and identified by Dr. K. Gauthaman of Pharmacognosy Department, Himalayan Pharmacy Institute, Sikkim, India. A voucher specimen number HPI 168 was deposited in the departmental herbarium.

**Extraction:** The whole plant was dried in shade and powdered (no. 60 mesh) and 100 g of the dried powder was extracted successively with petroleum ether, chloroform and methanol in Soxhlet apparatus. The weight of methanolic extract after drying was calculated as 15.4 g.

**Animals:** Male Sprague-Dawley rats weighing 300-350 g were used for the experiment. The rats were kept in suitable environmental conditions and fed with standard diet and water *ad libitum*. The experiment was carried out in accordance with the guidelines of Committee for the Purpose of Control and Supervision on Experiments on Animals (CPCSEA). The institute animal ethical committee has given approval for conducting animal experiments (HPI/08/60/IAEC/0060).

**Acute Toxicity Study:** The acute toxicity study of *Passiflora nepalensis* was performed. [9, 10] The dead animals obtained from primary screening studies, $LD_{50}$ value determination experiments, and the acute studies were subjected to post mortem studies. The external appearance of the dead animals, the appearance of the viscera, heart, lungs, stomach, intestine, liver, kidney, spleen and brain were carefully noted and any apparent and significant features or differences from the normal were recorded [1].

**Experimental Design for Hepatoprotective Activity:** Animals are divided into 5 groups, each comprising 6 rats.

*Group I:* Normal control (saline)
*Group II:* Simvastatin (20mg/kg p.o)
*Group III:* Simvastatin (20mg/kg p.o) + *Passiflora nepalensis* extract (150mg/kg, p.o)
*Group IV:* Simvastatin (20mg/kgp.o) + *Passiflora nepalensis* extract (225mg/kg, p.o)
*Group V:* Simvastatin (20mg/kgp.o) +Silymarin (25mg/kg, p.o)

Animals were divided into five different groups, each having 6 rats and treated accordingly. Group 1: rats fed with a normal standard diet for 30 days. Group II rats received Simvastatin (SMT) (20mg/kg p.o alone for 30 days).Groups III and IV rats received SMT along with *Passiflora nepalensis* extracts(150mg/kg and 225mg/kg p.o respectively for 30days) and Group V rats received SMT along with Silymarin (20 mg/kgp.ofor 30 days). On the 31st day, all the animals were sacrificed by mild ether anesthesia [11].

**Blood Biochemistry:** Blood samples were collected in glass tube from retroorbital puncture to obtain haemolysis free clear serum for the analysis of SGOT and SGPT [12], ALP [13] and Bilirubin [14] by standard method. Serum total protein was measured.[15]

**Statistical Analysis:** All the results were expressed as mean ± standard error (SEM). Data were analyzed using student'st-test. $p<0.001$ and $p<0.05$ were considered as statistically significant.

## RESULT

The effect of methanolic extract of *Passiflora nepalensis*on serum transaminases, alkaline phosphates, bilirubin and total protein level in Simvastatin intoxicated rats are shown in charts. There was a significant increase in bilirubin levels, SGOT, SGPT and ALP, in Simvastatin intoxicated group compared to the normal control group. The total protein levels were significantly decreased to 3.28g/dl in Simvastatin intoxicated rats from the level of 6.44 g/dl in normal group. On the other hand the groups with received both *Passiflora nepalensis* extract (150mg/kg and 225mg/kg,) + Simvastatin (20mg/kg p.o) (Groups III and IV) and Simvastatin (20mg/kg p.o) +Silymarin (25mg/kg, p.o) (Group V) showed significantly decreased the elevated serum marker enzymes when given orally and reversed the altered total protein to almost normal level.

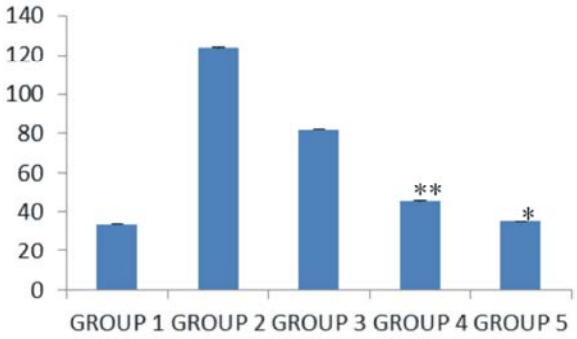


Fig. 1: Effect of methanolic extract of *Passiflora nepalensis* on SGPT levels in rats.

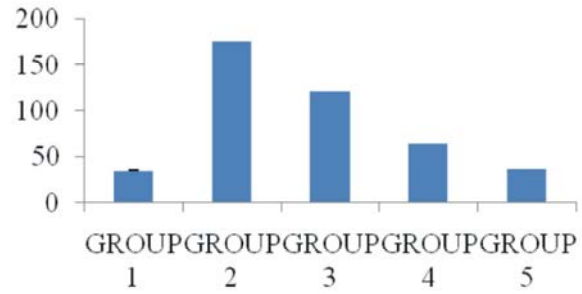


Fig. 2: Effect of methanolic extract of *Passiflora nepalensis* on SGOT levels in rats.

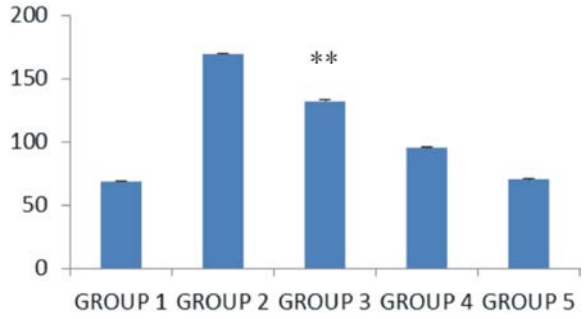


Fig. 3: Effect of methanolic extract of *Passiflora nepalensis* on ALP levels in rats.

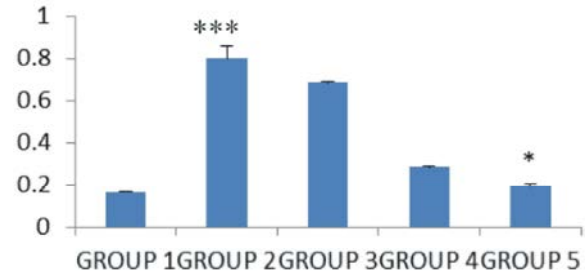


Fig. 4: Effect of methanolic extract of *Passiflora nepalensis* on Direct Bilirubin levels in rats.

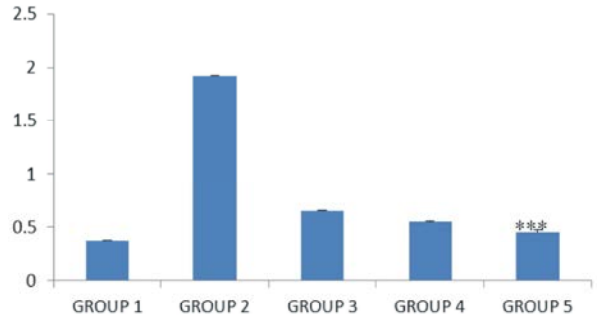


Fig. 5: Effect of methanolic extract of *Passiflora nepalensis* on Total Bilirubin levels in rats.

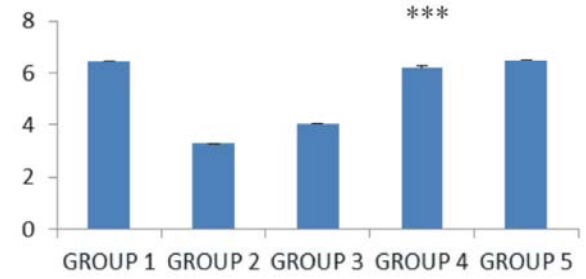


Fig. 6: Effect of methanolic extract of *Passiflora nepalensis* on Total Protein levels in rats.

## DISCUSSION

The liver can be injured by many chemicals and drugs. During hepatic damage, cellular enzyme like SGOT, SGPT, ALP and serum bilirubin present in the liver cell, leak into the serum resulting in increased concentration. This decrease in elevated serum levels followed by simvastatin-treated animals in part may be due to the protective effect of *Passiflora nepalensis* extracts on liver cells which causes the restoration of liver cell membrane permeability. It is already reported that simvastatin causes oxidative stress which leads to hepatotoxicity. The protection of liver cells against toxic materials including drugs, lipid peroxidation and free radical injury may decrease inflammation [16]. Histological changes such as steatosis (fatty changes in hepatocytes) and perivenular fibrosis were observed in simvastatin control group. Methanolic extracts of *Passiflora nepalensis* (150 mg/kg and 125mg/kg, p.o) prevented these histological changes.

## CONCLUSION

In conclusion, the results of present study demonstrate that *Passiflora nepalensis* extracts

(150 mg/kg and 225mg/kg,) has potent hepatoprotective activity against simvastatin induced liver damage in rats. The results also imply that the hepatoprotective effects of *Passiflora nepalensis* may be due to its antioxidant property. Further investigation is in progress to determine the exact phytoconstituents responsible for hepatoprotective effect.

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