

# Efficacy of leaf extract of *Agave americana* L. against carbon tetrachloride induced hepatic damage in albino rats

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## Abstract

Many hepatoprotective herbal preparations have been recommended in Indian systems of medicine for the treatment of hepatic disorders. Protective action of one such herbal preparation, viz., *Agave americana* leaf extract, was evaluated in animal mode (albino rats) with hepatotoxicity induced by carbon tetrachloride. Levels of bilirubin, alkaline phosphatase, serum glutamate oxaloacetate transaminase, serum glutamate pyruvate transaminase and cholesterol were significantly higher in hepatotoxicity induced rats. *Agave americana* leaf extract brought about recovery in the levels of all the above biochemical parameters suggesting its hepatoprotective potential.

**Keywords :** *Agave americana*, carbon tetrachloride, hepatoprotective activity, hepatotoxicity, liver injury

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## INTRODUCTION

Liver is involved in metabolic functions and is a frequent target of a number of toxicants (Meyer and Kulkarni, 2001). Carbon tetrachloride is an extensively studied liver toxicant, and its metabolites such as trichloro methyl peroxy radical ( $\text{CCl}_4\text{O}_2$ ) are known to be involved in liver damage (Mehandale *et al.*, 1987). It is a widely used chemical to induce liver damage in experimental studies and the resulting hepatic injury was characterized by leakage of cellular enzymes into the blood stream by centrilobular necrosis (Murael *et al.*, 2001). The involvement of free radicals in the pathogenesis of liver injury has been investigated for many years by using acute poisoning with  $\text{CCl}_4$  (Recknagel, 1989). The principle causes of carbon tetrachloride induced hepatic damage are lipid peroxidation, decreased activities of antioxidant enzymes and generation of free radicals (Poli, 1993). Enhancement of antioxidant activity and/or the inhibition of the generation of free radicals are important in providing protection against hepatic damage. There are a number of medicinal preparations in *ayurveda* (a traditional Indian system of medicine) recommended for the treatment of liver disorders (Chatterjee, 2000). One among them is the administration of leaf extract of *Agave americana* L. (Family: Amaryllidaceae), a medium to small sized tree distributed throughout India. The plant is also used as demulcent, laxative and antiseptic and for treating syphilis and tuberculosis (Kumar *et al.*, 2002). This paper evaluates the potential of the leaf extract of *A. americana* for hepatoprotective activity in albino rats with carbon tetrachloride induced hepatic damage.

## MATERIALS AND METHODS

### Preparation of extract

The leaves of *Agave americana* L. were locally collected in and around Tiruchirappalli, Tamil Nadu, South India. The plant was identified and authenticated with the Department of Botany, STET Women's College, Mannargudi, South India (STET 314).

Fifty grams of leaves were taken and ground well with distilled water using mortar and pestle. The herbal juice obtained was filtered and the filtrate was evaluated for hepatoprotective activity in albino rats with  $\text{CCl}_4$  induced hepatic damage.

### Experimental animals

Male albino rats of Sprague-Dawley strain weighing 120 – 150 g were secured from small animals breeding centre of King's Institute, Chennai, South India. They were housed in polypropylene cages maintained at controlled room temperature and normal light-dark cycle. The animals were fed with pellet diet and water *ad libitum*. A week's time was given for the animals to get acclimatized with the laboratory conditions.

### Experimental design

Animals were divided into four groups of six rats each as follows. Group I was the control. Group II animals received 1ml/kg b.w./day dose of  $\text{CCl}_4$  intraperitoneally for 21 days, group III animals received 30 mg/kg b.w./day oral dose of silymarin a standard drug used for treating liver disorders along with  $\text{CCl}_4$  for 21 days and the Group IV animals received 300 ml/kg b.w./day oral dose of leaf juice of *A. americana* along with  $\text{CCl}_4$  for 21 days. All the animals were sacrificed on the 22<sup>nd</sup> day. Blood samples were collected from the retro-orbital sinus and analysed for the activities of bilirubin, Alkaline Phosphatase (ALP) (E.C. No. 3.1.3.1), Serum

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Glutamate Oxaloacetate Transaminase (SGOT) (E.C. No. 2.6.1.1), Serum Glutamate Pyruvate Transaminase (SGPT) (E.C. No. 2.6.1.2) and cholesterol.

#### Biochemical estimation

Estimation of bilirubin, alkaline phosphatase (ALP), serum glutamate oxaloacetate transaminase (SGOT), serum glutamate pyruvate transaminase (SGPT) and cholesterol were carried out by the methods described by Malloy and Evelyn (1937), Bessay *et al.* (1964), Reitzman and Frankel (1957) and Allain (1974), respectively.

#### Statistical analysis

Results of biochemical parameters of liver were reported as mean  $\pm$  S.E. of six animals in each group. Significant intergroup differences were determined statistically by subjecting the data to ANOVA and values with  $P < 0.05$  were considered to be significant.

### RESULTS AND DISCUSSION

Levels of all biochemical parameters got significantly increased in group II rats after CCl<sub>4</sub> administration ( $P < 0.05$ ) as compared to control (Group I) (Table 1). The increased activities of enzymes such as SGOT, SGPT, ALP in the serum of CCl<sub>4</sub> induced rats indicate damage to hepatic cells (Jayatilaka, 1990 and Ohta *et al.*, 1995). Damage to the cell integrity of the liver by CCl<sub>4</sub> might have resulted in an increase in the activity of SGOT, which is released into circulation after cellular damage (Bandyopadhyay *et al.*, 1999). The *A. americana* leaf extract treatment resulted in significant decrease in the activities of all these enzymes i.e., reverted the effects of

CCl<sub>4</sub> treatment significantly (Table 1). ALP is a membrane bound enzyme and is released unequally depending on the pathological conditions. Effective control of its activity is indicative of improvement in the secretory mechanism of hepatic cells (Plaa and Zimmermann, 1997). Thus the *A. americana* leaf juice seemed to give protection against liver injury by CCl<sub>4</sub>.

Liver diseases are known to be associated with impaired hepatic drug metabolizing capacity and impaired activity of various hepatic enzymes (Wolf, 1999). Increased serum total bilirubin level in CCl<sub>4</sub> treated rats (vide Table 1) confirms the intensity of jaundice. In such toxic hepatitis, the degree of excretion of bilirubin from the intestine is very less and bilirubin present in the liver is excreted into canaliculi and regurgitated in to the blood stream. Leaf extracts of *A. americana* has significantly reverted the damage caused by CCl<sub>4</sub> by bringing back the serum bilirubin levels to near normal conditions (vide Table 1).

Marked alteration in lipid metabolism has been reported earlier in chronic CCl<sub>4</sub> administration (Day *et al.*, 1984). Our results also showed increased level of cholesterol in CCl<sub>4</sub> treated rats. The liver plays an important role in the synthesis and removal of cholesterol from the circulation. Disease status might result in bile duct obstruction leading to increased serum cholesterol level (Day *et al.*, 1984). The cholesterol level tends to rise in condition with either extra-hepatic (or) intra-hepatic injury and biliary obstruction and some times rises to very high levels in patients with primary biliary cirrhosis due to CCl<sub>4</sub> toxicity. The juice of *A. americana*

Table 1. Effect of *Agave americana* leaf extract on the levels of serum bilirubin, Alkaline Phosphatase (ALP), Serum Glutamate Oxaloacetate Transaminase (SGOT), Serum Glutamate Pyruvate Transaminase (SGPT) and cholesterol against carbon tetrachloride induced hepatotoxicity in albino rats

	Bilirubin (mg / dl)	ALP (IU/l)	SGOT (IU/l)	SGPT (IU/l)	Cholesterol (mg / dl)
Group I (Control)	0.94 $\pm$ 0.007*	0.26 $\pm$ 0.008*	84.8 $\pm$ 0.04*	70.4 $\pm$ 0.19*	80.0 $\pm$ 0.01*
Group II (CCl <sub>4</sub> treated)	2.76 $\pm$ 0.009	0.409 $\pm$ 0.001	119.4 $\pm$ 0.05	152.7 $\pm$ 0.05	99.6 $\pm$ 0.05
Group III (Silymarin treated)	0.10 $\pm$ 0.06*	0.23 $\pm$ 0.0009*	95.2 $\pm$ 0.06*	70.8 $\pm$ 0.03*	84.2 $\pm$ 1.67*
Group IV ( <i>A. americana</i> leaf juice treated)	0.99 $\pm$ 0.02*	0.27 $\pm$ 0.004*	87.8 $\pm$ 0.03*	71.2 $\pm$ 0.08*	80.5 $\pm$ 0.20*

Values are mean  $\pm$  S.E. for 6 animals in each group.

\* $P < 0.05$  as compared with CCl<sub>4</sub> induced group

leaves reduced the cholesterol level significantly which indicated its effectiveness in hepatoprotection.

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