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Hepatoprotective Effect of Cardiospermum Canescens on Carbon Tetrachloride Induced Hepatotoxicity in Albino Rats.

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ABSTRACT

The different extracts of *Cardiospermum canescens(Sapindaceae)* were tested for their hepatoprotective activity against CCl_4 induced hepatotoxicity in albino rats. The degree of protection was measured by using biochemical parameters like serum transaminases, alkaline phosphatase, total protein and albumin. The methanolic extract showed the most significant hepatoprotective activity comparable with standard drug Silymarin. Other extracts namely petroleum ether and acetone also exhibited a potent activity.

KEY WORDS: Cardiosperum caneses, Methanol, Carbon tetrachloride, Silymarin.

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INTRODUCTION:

Cardiospermum canescens, belongs to the family Sapindaceae. The genus Cardiospermum comprises of two species. It is locally known as *pedda budda* in Telugu language. The whole plant has different properties like diaphoretic, diuretic, emetic, emmenagogue, laxative, refrigerant, rubefacient and stomachic. It is used in the treatment of rheumatism, nervous diseases, stiffness of the limbs and snakebite. Leaf juice mixed with cumin is consumed to relieve pain in the joints and given at the time of delivery. The roots are diuretic and used in treating liver disorders and dysentery. The leaf juice is used for the treatment of asthma . The leaves are rubefacient and are applied as a poultice in the treatment of rheumatism [1,2,3]

Liver plays a very important role in the metabolism of foreign compounds entering the body. The exposure to the foreign compounds may be through consumption of alien/contaminated foods from exposure to chemical substances in the occupation environment or through synthetic drugs consumed for various pathological conditions; these compounds have many toxic effects on the human liver. The liver gets injured also by viruses, chemicals, alcohol and autoimmune diseases. Liver diseases remained one of the serious health problems, so medicinal plants and herbs have been use for treating such problems as in the Indian traditional systems of medicine, especially Ayurveda. Recently, a scientific basis was proved to justify the various medicinal uses of herbs. India is well-known for a plethora of medicinal plants. The medicinal use of many plants (as hepatoprotectants) like *Andrographis paniculata, Azardiracta indica, Cassia fustula, Elephantopus scaber, Hibiscus rosasinensis, Phyllanthus debilis, Phyllanthus nesseri, Picrorrhiza kurroa, Cleom Viscosa, Annona squamosa, Ficus benjamena, Borreria articularis* and *Glycyrrhiza glabra* has been reported in the literature [4,5,6,7].

MATERIALS AND METHOD:

The roots of *Cardiosperum caneses* were collected in the month of August from Thirumala hills, Thirupathi, Andhra Pradesh, India, and authenticated by Dr. Madhavashetty, Professor, Department of Botany, Sri Venkateshwara University, Thirupathi, Andhra Pradesh, India. Voucher specimens are being maintained in the herbarium of Talla Padmavathi Pharmacy College, Jawaharlal Nehru Technological University, Hyderabad, Andhra Pradesh, India.

Preparation of extracts

Roots of *Cardiosperum caneses* were cleaned from any adherent foreign material and air-dried and crushed to powder and then successfully extracted to exhaustation with different solvents like petroleum ether, chloroform, methanol using Soxhlet apparatus. The different extracts those obtained were dried under reduced pressure to get the crude extract.

Animals

Female albino Wistar rats weighing 150 to 200 grams were purchased from Mahaveera Agencies, (Regn. No. 146/1999/CPCSEA), Hyderabad and maintained in the animal house of Talla Padmavathi College of Pharmacy, Warangal. Animals were provided with standard rodent pellet diet and the food was reserved 18 to 24 hr before the experiment, while water was allowed *ad libutum*. They were maintained at (25 ± 2°C) 12 hr light and dark cycle throughout the period of acclimatization and experimentation. All the animal experimental protocols were duly approved by the Institutional Animal Ethics Committee (Reg No.169/1999/UCPSc, KU).

Phytochemical analysis

Phytochemical tests were carried out to identify the phytoconstituents, such as carbohydrates, Amino acids, proteins, alkaloids, steroids, flavonoids, triterpenoids and saponin .

ACUTE TOXICITY STUDY

Wistar albino mice of 25 to 30 g were divided into ten groups of six animals each. Acute toxicity study was carried out according to the method described (Palanichamy and Nagarajan, 1990). The extract of *Cardiosperum caneses* were suspended in 5% gum acacia in doses of 100, 200, 400, 600, 800, 1000, 1200, 1400, 1800 and 2000 mg/kg and were given orally to albino mice. The animals were observed continuously for any change in autonomic or behavioral responses for first few hours and later at 24 h intervals for a period of 48 h. At the end of this period, the mortality rates in all groups were noted.[18]

HEPATOPROTECTIVE ACTIVITY

In the present study, the animals were pretreated with test extract before inducing liver damage with CCl_4. Seven days after acclimatization, the rats were divided into five groups (1 to 5) each group consisting of six animals. All animals were kept on same diet for 7 days.

Group I: Served as a normal group and received 1 mg/kg of 2% w/v gum acacia in water for few days.

Group II: Treated with vehicle (1 mg/kg of 2% w/v gum acacia in water) daily for seven days followed by CCl_4 on the seventh day.

Group III (standard-Silymarin): Animals received 50mg/kg of Silymarin for seven days orally followed by CCl_4.

Group IV: This group were treated in the similar way using petroleum ether of 250mg/kg, respectively followed by CCl_4 administered orally.

Group V: This group were treated with chloroform of 250mg/kg, respectively followed by CCl_4 administered orally.

Group VI: This group were treated with methanolic extract of 250mg/kg , respectively followed by CCl_4 orally.

Assessment of liver function

Rats of all groups were anaesthetized by diethyl ether 24 hr after the administration of hepatotoxin like CCl_4. The blood was obtained from all groups of rats by puncturing retro-orbital plexus. The blood samples were allowed to clot for 45 min at room temperature. Serum was separated by centrifugation at 2500 rpm at 30 °C for 15 min and analyzed for various biochemical parameters: Serum transaminases, Serum glutamic oxalo transaminase (SGOT), Serum glutamic pyruvic transaminase (SGPT), Alkaline phosphatase (SALP), Total protein (TP) and Total albumin (TA) according to the reported methods [8,9,10].

Statistical analysis

Results of the biochemical estimations are reported as mean S.E.M. Total variation, present in a set of data was estimated by one-way analysis of variance (ANOVA), Student's *t*-test was used for determining significance [11].

RESULT AND DISCUSSION

Administration of CCl_4 led to increase the serum enzymes level by 2–3-folds as compared to control group. Treatment of rats with different extracts of *Cardiospermum canescens* at dose of

250mg/kg b.w. p.o. markedly prevented CCl_4 induced elevation of SGOT, GPT, SALP and increased the level of TP and TA Table.1

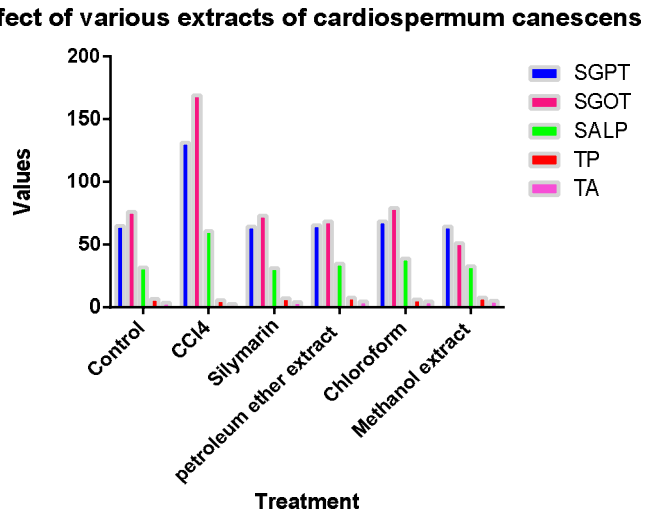
Table.1 Effect of various extracts of roots on serum enzymes alkaline phosphatase, total proteins and albumin in CCl4 induced in liver damage in rats.

Treatment	SGPT	SGOT	SALP	TP	TA
Control	65.1±0.2	76.1±3.05	31.66±1.28	6.99±0.11	3.82±0.20
CCl4	131.49±1.86	169.24±4.56	61.01±0.89	5.50±0.06	2.70±0.01
Silymarin	64.31±1.01*	73.28±3.30*	31.21±1.19*	7.20±0.31*	4.10±0.01
Petroleum ether extract	65.39±0.99*	68.76±1.05	34.59±0.49	7.70±0.10	4.61±0.09
Chloroform	68.79±5.0*	79.29±4.72*	38.90±1.19	6.32±0.49	4.51±0.09
Methanol extract	64.31±1.25*	51.21±9.53*	32.58±0.77*	7.9±0.25*	5.20±0.15*

Values are mean±SEM., n=6 animals per each group. The percentage of protection in calculated as 100x (values of CCl_4 control –values of sample)/(values of CCl_4 control- values of control). SGOT- Serum glutamic oxaloacetic transaminase; SGPT- Serum glutamic pyruvic transaminse; SALP, alkaline phosphatase; TP- total protein; TA- total albumin;

*P<0.05,

**P<0.01 vs CCl_4. One way analysis and student's t-test.



CCl_4 induces fatty liver and cell necrosis and plays a significant role in inducing triacylglyceral accumulation, increased lipid peroxidation, membrane damage, depression of protein synthesis and loss of enzymes activity . Being cytoplasmic in location the damage marker enzymes SGOT, SGPT and LDH are released in serum [12,13,14,15]. It has been shown that protective agents exert their action against CCl_4 induced liver injury by impairment of CCl_4 mediated lipid peroxidation, either through decreased production of free radical derivatives or due to the antioxidant activity of the protective agent itself [16,17]. The extracts of the roots of *Cardiospermum canescens* used in the study preserved the structural integrity of the hepatocellular membrane in a dose dependent manner as evident from the protection provided as compared to the enzyme levels in CCl_4 treated rats.

Furthermore, protective mechanism not specific to CCl_4 may be responsible for hepatoprotective activity of the methanolic extract of the roots of *Cardiospermum canescens*

CONCLUSION

The above observations have shown that the methanolic extract of the roots of *Cardiospermum canescens* showed maximum antihepatotoxic activity, which should be related to the methanol soluble active principles like flavone and diterpene whereas, the other extracts of roots of *Cardiospermum canescens* also showed a lower antihepatotoxicitivity. The activity of the tested samples was comparable to that of standard drug Silymarin. The isolation and testing of constituents likely to be responsible for the hepatoprotective activity of *Cardiospermum canescens.*

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