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HEPATO-PROTECTIVE ROLE OF FLOWER EXTRACT OF COUROUPITAGUIANENSIS AUBL AGAINST CHLORAMPHENICOL INDUCED HEPATO-TOXICITY IN MUSMUSCULUS

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ABSTRACT:

The medicinal plants have been used traditionally by the physician worldwide for the prevention and treatment of various liver disorders with no ill effects. The plant kingdom have main role in the life of human beings as well as animals. Considerable studies have been carried out to assess the hepato-protective activities. The clinical research in this century has confirmed the efficacy of several medicinal plants in the treatment of liver disease.

The flower extract of *Couroupitaguianensis*(FE of CG) was evaluated for its protective role against chloramphenicol induced hepato-toxicity in mice. The activities of serum marker enzymes of liver injury like serum glutamate pyruvate transaminase (SGPT), serum glutamate oxaloacetate transaminase (SGOT), alkaline phosphatase (ALP), which are increased by chloramphenicol treatment was found to be reduced by the treatment of flower extract of *Couroupitaguianensis*at 250 mg/kg body weight except SGPT which was increased. The lipid peroxidation in liver, the marker of membrane damage was found to be nearly normal level and bilirubin content in serum was found to be at significantly low level in the extractgroup, indicating its protective role. The treatment with extract produced enhancement of antioxidants enzymes like catalase (CAT) and reduced glutathione (GSH). The result suggest that the protective role of the flower extract of *Couroupitaguianensis*against chloramphenicol induced hepato-toxicity. The phytochemicals estimated from the plant was swietenine, sapropterin, usnic acid, lupeol and gamma tocopherol. The possible mechanism of action of flower extract may be due to its antioxidant activity and free radicals.

Keyword: Chloramphenicol, Couroupitaguianensis, antioxidants, free radicals, hepato-protection.

I. INTRODUCTION

Chloramphenicol is an antibiotic derived from the bacterium Streptomyces venezuelae or produced artificially and effective against a broad spectrum of micro-organisms. Chloramphenicol is a highly effective and well tolerated broad–spectrum antibiotic. However, it has several features that demand careful use in companion animals and that have led to prohibition of its use in food producing animals in several countries, including USA and Canada (Kahn, 2005).

Chloramphenicol is excessively used in the developing and undeveloped countries against the microbial infections because it is cheap and effective. However, the high dose of chloramphenicol causes liver-toxicity due to formation of free radicals or reactive oxygen species (ROS). Free radicals produce deleterious effect on lipid plasma membrane as well as cellular components thereby producing peroxidation of lipids which leads to cell death (Ryter et al., 2007).

Medicinal plants possess scavenging activity for free radicals and boost the antioxidant defence mechanism in body and have a protective role against tissue damage induced by chloramphenicol (Kumar K B H and Kuttan R, 2005). *Couroupitaguianensis*Aubl. was the selected medicinal plant for the research study. Many parts of *Couroupitaguianensis*have been used traditionally to treat various diseases, like the decoction of its flowers is used to boost the immune system for fighting a number of diseases (Kokate, C. K., 1988).The

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extracts of leaves and flowers are used as microbial activities (Al. Dhabi, N. A. et. al., 2012). It is used extensively as an ingredient in many preparations which cure gastritis, scabies, bleeding piles, dysentery, scorpion poison and the flowers of *C. guianensis*showed analgesic and anti-inflammatory activity and immunomodulatory activity (Geeta M, et al., 2005, Pradhan D, et al., 2008 and FarrukhAqil, et al., 2006).

II.MATERIALS AND METHODS

I Plant material and authentication

The flowers of plants collected from local region of Mumbai and the plant is authenticated by BLATTER HERBARIUM, ST. Xavier's college, Mumbai-400001, India.

II Preparation of plant extract

The Flower powdered was first extracted with petroleum ether (60-80°C) to remove the fatty contents and the extract was discarded. The residue was exhaustively extracted in a soxhlet apparatus for at least 12 hour with methanol and the extract was used for experiment. The solvent from extract was removed under reduced pressure controlled temperature (40°-50°c). The yield of methanolic extract was approximately 16/17 % w/w. The dried semisolid extract was kept in lightly closed container in refrigerator till further analysis. (Vinod H. Gupta, et al., 2012).

III Animals- mice

The animals used for the studies of toxicity and for efficacy were healthy Albino Swiss mice (Musmusculus), weighing between 30-35 gm obtained from Haffkins Institute, Parel (E), Mumbai- 400012. Under the Animal Maintenance permit Registration Number Invochem Laboratory, 226, "Gauri" Commercial Complex, Station Road, Vasai Road (E), Dist. Thane-401210; CPCSEA Registration No. 851/C/04/CPCSEA, from the ministry of Social Justice and Empowerment, Government of India. After procurement, the male and female mice were kept in same cage. The cages were provided with rice husk bedding and were cleaned daily. The house was maintained at 28±2° c and exposed to 10-12 hours of day light and a relative humidity of 30-70 %. The animals were provided with drinking water ad libitum and fed on commercially available feed supplied by AMRUT FEED.

IV Drug- chloramphenicol

Chloramphenicol was procured from Mehta Pharmaceutical Limited, 315, Janki Centre, Plot No. 29, Shah Industrial Estate, off Veera Desai road, Andheri (W), Mumbai, India. It is kept in below room temperature. Chloramphenicol is beneficial to control the growth of gram positive and gram negative bacteria, however chloramphenicol at high concentrations results in hematotoxicity, linkage to fatal aplastic anaemia (Saba et al., 2000) **9** and hepatotoxicity. LD₅₀ of chloramphenicol is 2300 mg/kg body weight of mouse according to Pfizer material safety data sheet, 2007.

V Experimental protocol

Group I (6 mice) were used as controls. Group II (6 mice) received chloramphenicol i.e. 500 mg/kg. Group III (6 mice) received 200 mg of flower extract of *Couroupitaguianensis*. Group IV (6 mice) received chloramphenicol i.e. 500 mg/kg and 200 mg/kg of flower extract of *Couroupitaguianensis*.

VI Blood sample collection and analysis

Blood sample was collected by puncture of retro- orbital vein and put the blood in EDTA vial for all hepatological analysis like SGPT, SGOT, ALP, superoxide dismutase (SOD), catalase (CAT), reduced glutathione (GSH) and lipid peroxidase (LPO).

Statistical analysis

The obtained DATA was expressed as mean \pm SD. Statistical significance of differences between the control and experimental groups was assessed by Analysis of variance (ANOVA) two ways without replication. The value of probability less than 5 % (P<0.05) was considered statistically significant.

III.RESULTS

Table 1 show there was significant increase in hepatic marker enzymes like SGPT, SGOT, ALP in all groups as compare to control group except ALP was lower than control group. While table 2 showsthere was

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improvement of CAT and GSH in prophylactic group, while SOD and LPO werenot improved in flower extract as well as prophylactic groups.

IV.DISCUSSION

Assessment of liver damage can be made by estimating the activities of serum enzymes SGPT, SGOT, ALP which are originally present in higher concentration in cytoplasm. When there is hepatopathy, the enzymes leak into the blood stream in conformation with the extent of liver damage (Tanaka K and Lizuka Y; 1968). The increased level of these marker enzymes observed in the toxin group i.e. chloramphenicol treated mice in the present study correspond to the more liver damage induced by toxin. The reduced concentrations of SGOT and ALP as a result of flower extract of *Couroupitaguianensis*Aubl administered to mice thereby decreasing enzyme linkage. It may be due to presence of swietenine, sapropterin, usnic acid, lupeol and gamma tocopherol indicating hepato-protective potential of flower extract of CouroupitaquianensisAubl.

It is suggested that swietenine, sapropterin, usnic acid, lupeol and gamma tocopherol in the flower extract of CouroupitaquianensisAubl. play an important role as antioxidant for prevention of hepatic damage. These phytocompounds of flower extract of *Couroupitaguianensis*Aubl. may able to stabilise ROS by reacting with them and oxidizes subsequently to more stable and less reactive radicals.

As flower extract of Couroupitaquianensishas swietenine, sapropterin, usnic acid, lupeol and gamma tocopherol which have extra electron, it shared with free radicals which already have one electron less. So the plant extract recovered the toxicity effect which is caused by chloramphenicol. However SGPT, SOD and LPO values were not decreased, so the damaged liver was not recovered completely.

CouroupitaguianensisAubl in mice.							
Groups	SGOT IU/L	SGPT IU/L	ALP IU/L	SOD U/mg	CAT OD/mg	GSH ug/mg	LPO nmoles/gm
I- Control	178±16.4	45±6.03	249 ± 43.61	35.6±7.23	3.8±0.44	4.78±0.77	117.27±2.66
II- Chloram	194±46.38	47.67 ± 4.92	292 ± 39.86	24.9±5.71	1.93±0.78	2.88±0.28	161.54±15.85
III- FE of CG	154 ± 13.5	53 ± 6.9	238 ± 56.9	24.5±7.98	3.71±0.54	3.76±0.73	125.45±8.71
IV-Chloram + FE of CG	157 ± 20	54±5.43	313 ± 92	21.3±5.31	2.6±0.028	3.29±0.52	171.71±21.49

TABLE-1

Hepatological observations after treatment and recovery with the help of flower extract of

values< 0.05 by 'f' test. The values are expressed as Mean ± SE from 6 rats in each group. FE of CG means flower extract of Couroupitaguianensis

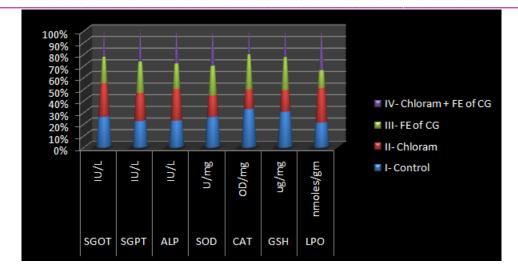


Fig. 1 – Effect of flower extract of *Couroupitaguainensis* on liver function markers of mice with chloramphenicol – induced hepatological changes (values are mean \pm SE from 6 mice per group. P values: < 0.05; compared normal control group, chloramphenicol group 500 mg/kg body wt, recovery group i.e. flower extract of *Couroupitaguainensis* (200 mg/body wt/day) group and chloramphenicol 500 mg/kg body wt with flower extract of *Couroupitaguainensis* (200 mg/body wt/day) group).

V.CONCLUSION

The biochemical evidences show that the treatment of flower extract of *Couroupitaguianensis*protected mice moderately against chloramphenicol induced hepato-toxicity. The phytoconstituents of the flower extract of *Couroupitaguianensis*have active antioxidant role toprotect the liver from LPO as well as marker enzymes. The further studies should be conducted to know the role of phytochemicals to protect the other organs also.

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