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Short Communication

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Hepatoprotective activity of Herbadict tablet: An ayurvedic herb mineral product on alcohol induced mortality and liver lesions in mice

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Abstract

The main objective is to study the possible protective effects of Herbadict tablet, a polyherbal ayurvedic product against alcohol-induced mortality and liver lesions in mice. Healthy female Swiss albino mice were divided into six groups containing 8 animals each. Herbadict tablet was suspended in 0.5% Carboxy Methyl Cellulose (CMC) and administered at two doses i.e., 30 mg/kg and 100 mg/kg body weight. Vehicle control, positive control and reference control (Silymarin) were used for comparison. Alcoholic intoxication was induced using ethanol. Treatment with Herbadict significantly ($p<0.01$) decreased SGOT, SGPT and -GT at 100 mg/kg when compared to the vehicle treated alcohol intoxicated group. A dose dependent significant decrease in total bilirubin was observed at 30 and 100 mg/kg of Herbadict when compared to vehicle treated alcohol intoxicated group. Treatment with Herbadict at 30 and 100 mg/kg p.o. showed hepatoprotective effect against alcohol induced damage in mice model.

Introduction

Alcoholic intoxication was induced using ethanol. Concentration of ethanol was progressively increased in the drinking water viz 10% (v/v) alcohol in the first week, 20% in the second, 30% in the third, and 40% in the fourth week. Animals were treated with vehicle or test item simultaneously (10.00 am to 11.00 am) every for 28 days. On day 29, the animals were fasted for 4 h and blood samples were collected for biochemical analysis. The animals were then euthanized (ketamine 100 mg/kg, i.p) and liver sample were harvested for histopathological evaluation.

Parameters -

Liver function test Plasma Biochemistry - ALT, AST, ALP, γ-GT and total bilirubin using diagnostic kit (Spinreact, Spain) in Semiautomatic biochemical analyzer (Labmate, India).

Histopathology: Liver collected from mice were fixed in 10% neutral buffered formalin solution, dehydrated in graded alcohol and embedded in paraffin. Paraffin sections of 3-5 micron were mounted on glass slides and counter-stained with Hematoxylin and Eosin (H&E) for light microscopic analyses.

Data analysis; Data were expressed as mean ± SEM. Mean difference between the groups were analysed by One way anova followed by Tukey's multiple comparison test as post hoc. P value 0.05 was considered as statistically significant.

Ethical clearance: Approval was taken from Institutional Animal Ethical Committee before commencing the study

Acclimatization and Grouping: Female Swiss albino mice aged between 9 to 10 weeks were acclimatized for six days under laboratory conditions. Randomization was performed on last day of acclimatization. Mice were maintained at ideal macro and micro environment, standard laboratory diet and reverse osmosis water ad libitum. Healthy mice were selected and grouped based on the stratified body weight. Animals were grouped as under: Animals were divided into six groups containing 8 animals.

Dose formulation: Test item was suspended in 0.5% carboxy methyl cellulose (CMC). Test item was freshly prepared in vehicle prior to dosing. Test item was weighed, transferred to mortar and grinded with pestle. A small quantity of the vehicle was added to test item and triturated. This was transferred to a measuring cylinder. A small quantity of the vehicle was added to motor again, triturated and transferred to the measuring cylinder. Sufficient quantity of vehicle was added to make up the required volume of formulation. The formulation prepared was then transferred to motor and again triturated. Following trituration, the formulation was transferred to a labelled beaker for dosing. The dose volume of the formulation was 10 ml/kg body weight.

Results and Discussion

Plasma Biochemistry (Figures 1-5)-Liver function test: Alcohol intoxicated mice showed significant ($p<0.01$) increase in SGOT, SGPT, ALP, γ-GT and total bilirubin levels when compared to vehicle control mice. Treatment with Herbadict significantly ($p<0.01$) decreased SGOT, SGPT and -GT at 100 mg/kg when compared to the vehicle treated alcohol intoxicated group. A significant ($p<0.01$) decrease in ALP was recorded at both 30 and 100 mg/kg doses of Herbadict when compared to the vehicle treated alcohol intoxicated group. A dose dependant significant decrease in total bilirubin was observed at 30 and 100 mg/kg of Herbadict when compared to vehicle treated alcohol intoxicated group. Effect of Herbadict was comparable with that of the reference drug, Silymarin

Histopathology

Group I: Vehicle control: Liver section showed normal architecture with central vein, hepatocytes radiating from the central veins and the portal triads. •

Group II: Positive Control: Liver sections revealed moderate to severe degree of hepatocellular vacuolations multifocal areas of ballooning degeneration of the hepatocytes. Few hepatocytes showed granular cytoplasm. Dilatation of sinusoidal spaces containing erythrocytes was also noticed. Focal areas of perivascular lymphocytic infiltration and centrilobular necrosis •

Group III: Reference control: Liver sections revealed moderate degree of vacuolations within the hepatocytes, mild degree of ballooning degeneration in hepatocytes and mononuclear cells infiltration in the necrotized area. •

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Group IV: Treatment with Herbadict at 30 mg/kg p.o. Liver sections revealed mild degree of hepatocellular vacuolations, minimal degree of ballooning degeneration, absence of sinusoidal dilatation and necrosis. Focal area of restoration of normal parenchyma was evident. Group V: Pre-treatment with Herbadict at 100 mg/kg p.o.: Liver sections revealed moderate degree of vacuolations and ballooning degeneration of hepatocytes, focal areas of perivascular infiltration and centrilobular necrosis. • Group VI: Pre-treatment with Livokot at 300 mg/kg p.o.: Liver lesions were similar to the positive control.

Test product contains the herbs having hepatoprotective and antioxidant activities. In vivo hepatoprotective effect of aqueous extract of Phyllanthus niruri on nimesulide-induced oxidative stress is established4 . Vinay et.al, in their work have established the Hepatoprotective activity of combination of Phyllanthus niruri and Curcuma longa extracts against ethanol induced toxicity in wistar rats5 . Harish R and Shivanandappa T. have proved antioxidant activity and hepatoprotective potential of phyllanthus niruri6 . Picroliv active constituent of Picrorhiza kurrooa7 , is found to have protective activity against oxytetracycline induced hepatic damage. Rawat A.K. et.al. In their study have established hepatoprotective activity of Boerhaavia diffusa L. roots8 . Hepatoprotective activity of Eclipta alba hassk against paracetamol induced hepatocellular damage9 and carbon tetrachloride induced acute liver damage10 are reported. Aqueous extract of Tinospora cordifolia has shown encouraging hepatoprotective activity11 . Summative effect of these multiple ingredients might have contributed to the hepatoprotective activity of the product