Pre-clinical evaluation of hepatoprotective activity of phytol in wistar albino rats

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Abstract

Introduction: Phytol, a phytoconstituent has been attributed for its hepatoprotective effect compared to Silymarin and hence present study.
Aim: To study hepatoprotective activity of phytoconstituent Phytol in Ethanol induced experimental liver injury in rats.
Materials and Methods: Wistar rats (male and female equal number) were divided into 5 groups consisting of 6 animals per group. The animals were pre-treated twice daily with 100 mg/kg and 200 mg/kg of Phytol which served as test group and Silymarin 100mg/kg orally which served as standard 1 hour before ethanol administration. All the animals except normal group animals were received ethanol (3.76 g/kg) twice daily for a period of 25 days. On 26th day, the animals were sacrificed to collect the sample to analyse levels of serum glutamic pyruvic transaminase (SGPT), alkaline phosphatase (ALP), serum glutamic oxaloacetic transaminase (SGOT), , cholesterol, triglyceride (TG), bilirubin, total protein (TP), super oxide dismutase (SOD), catalase (CAT) and glutathione (GSH). Phytol statistically reduced increased levels of SGPT, SGOT, ALP, TG, cholesterol and bilirubin in diseased animals. Phytol statistically increased reduced levels of TP, SOD, CAT, GSH in diseased animals. It was concluded that the phytoconstituent phytol have hepatoprotective activity. Statistical analysis
Statistical Analysis: It was done using Graph Pad Prism version 4 software (Graph Pad Inc., USA). ANOVA followed by Bonferroni's Multiple Comparison test was applied. Data presented as MEAN±SEM. Confidence level was taken at 95%.
Results: The present study showed both low (100 mg/kg) and high (200 mg/kg) doses of phytol showed statistically significant (<0.001) hepatoprotective activity in the ethanol induced pharmacological animal model.
Conclusion: Phytol, a phytoconstituent can be used as hepatoprotective agent as an alternate to Silymarin.

Keywords: Hepato-protective, Pre-clinical evaluation, Phytol, Silymarin.

Introduction

Liver is an important organ in our body.[1] It carries out important functions in our body such as like removing toxins, neutralising toxins and removing harmful substances. Liver failure is a life threatening condition. According to WHO report, 60000 persons die of inflammation of liver and more than 170 million people suffer from long term liver infections. Now a day more and more persons consume excess alcohol and overloads the body with allopathic drugs. All these things lead to liver injury. Allopathic medicines offer no sufficient hepatoprotection and produce various side effects which varies from simple nausea and vomiting up to life threatening condition.[2] Hence there is a need to develop alternative medicines to treat and prevent hepatic disorders. Herbals are safe, well tolerated and free from harmful side effects.[3] Phytol is an herbal phytochemical phytoconstituent. It is widely spread in nature in all the plants. It is an acyclic diterpene.[4] As of today update o recorded data are available regarding the effect of phytol on liver damage, hence following study was undertaken.

Materials and Methods

Chemicals

Phytol and Silymarin were purchased from Yucca Enterprises- Mumbai. Ethanol was purchased from J Enterprises- Chennai. All chemicals, solvents used for this study were of the analytical grade obtained from Merck Specialties Private Limited- Mumbai, RFCL Limited- New Delhi, Finar Chemicals Limited- Ahmadabad, India. Biochemical estimation kits were procured from Robonik India Pvt. Limited- Mumbai.

Animals

Wistar rats (150-250g) were weighed and selected for the study and were kept in polypropylene cages for the purpose of housing under 12 hours' dark-light cycle between 25±5^{o}C temperature with humidity 50±5%. Animals were on standard pellet diet with free access to drinking water *ad libitium*. Institutional Animals Ethics Committee (IAEC) approval was obtained before conducting the study with Ref No. SDCP/IAEC/04/2018.

Dose selection

Dose selection study was performed according to OECD 423 guidelines. Here 2 groups of animals (Swiss albino mice) were taken viz., 300mg/kg and 2000mg/kg groups. Test drug phytol was administered for 14 days according to the above doses. During this, the physical parameters like changes in fur, skin, respiratory, mucus membrane, respiratory, circulatory, ANS, CNS, behaviour and observations of tremors convulsions, salivations, diarrhoea, lethargy, sleep, coma were observed. After 14 days of study the test drug phytol failed to show any signs of toxicity as described above. Depending upon the study the lower and higher doses of phytol was fixed at 100mg/kg and 200mg/kg body weight.[5]

Ethanol Induced Hepatotoxicity

Procedure

All the animals except normal group animals were received ethanol (3.76 g/kg) twice daily for a period of 25 days. Animals were sacrificed on 26th day as per CPCSEA guidelines and sample collection of serum from blood and extraction of liver from body were done.[6,7]

Grouping

Table

Group	Drugs	No. of animals
A	Control	6
B	Diseased Ethanol control animals	6
C	Low dose Phytol + Ethanol	6
D	High dose Phytol + Ethanol	6
E	Standard drug Silymarin + Ethanol	6

Parameters

The parameters estimated were: Serum= Serum glutamic pyruvic transaminase (SGPT), serum glutamic oxaloacetic transaminase (SGOT), alkaline phosphatase (ALP), cholesterol, triglyceride (TG), bilirubin, total protein (TP). All these were estimated through kits from Robonik. Liver homogenate= Super oxide dismutase (SOD),[8] catalase (CAT),[9] glutathione (GSH).[10]

Statistical analysis

Statistical analysis was done using Graph Pad Prism version 4 software (Graph Pad Inc., USA). ANOVA followed by Bonferroni's Multiple Comparison test (compare all) was applied. Data presented as MEAN±SEM. Confidence level was taken at 95%.

Results

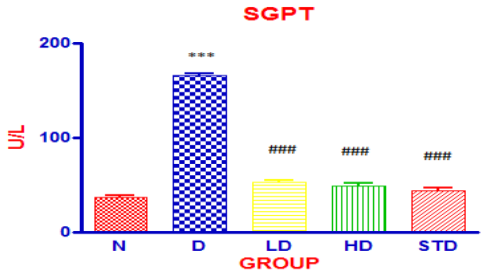

Fig. 1: Effect of phytol on SGPT. Values are expressed as MEAN±SEM. n=6.

*p	<0.05
** p	<0.01
*** p	p<0.001
#p	<0.05
##p	<0.01
### p	p<0.001

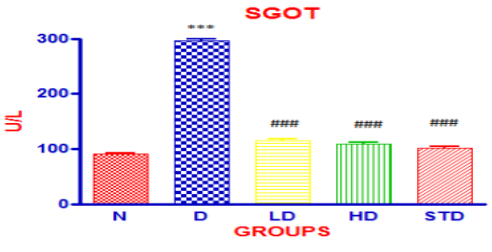

Fig. 2: Effect of phytol on SGOT. Values are expressed as MEAN±SEM. n=6.

*p	<0.05
** p	<0.01
*** p	p<0.001
#p	<0.05
##p	<0.01
### p	p<0.001

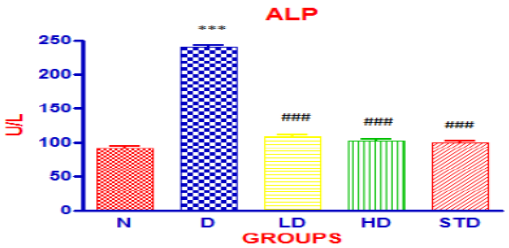

Fig. 3: Effect of phytol on ALP. Values are expressed as MEAN±SEM. n=6.

*p	<0.05
** p	<0.01
*** p	p<0.001
#p	<0.05
##p	<0.01
### p	p<0.001

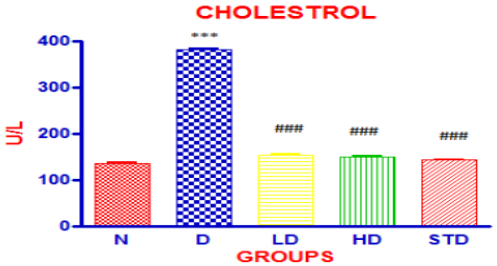

Fig. 4: Effect of phytol on Cholesterol. Values are expressed as MEAN±SEM. n=6.

*p	<0.05
** p	<0.01
*** p	p<0.001
#p	<0.05
##p	<0.01
### p	p<0.001

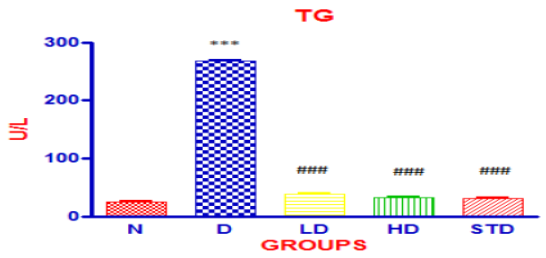

Fig. 5: Effect of phytol on TG. Values are expressed as MEAN±SEM. n=6.

*p	<0.05
** p	<0.01
*** p	p<0.001
#p	<0.05
##p	<0.01
### p	p<0.001

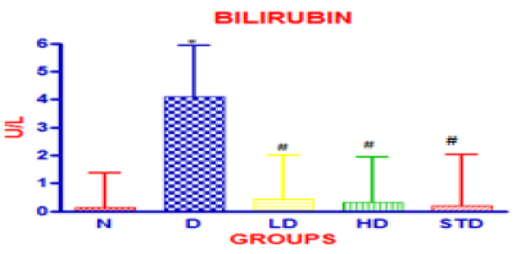

Fig. 6: Effect of phytol on Bilirubin. Values are expressed as MEAN±SEM. n=6.

*p	<0.05
** p	<0.01
*** p	p<0.001
#p	<0.05
##p	<0.01
### p	p<0.001

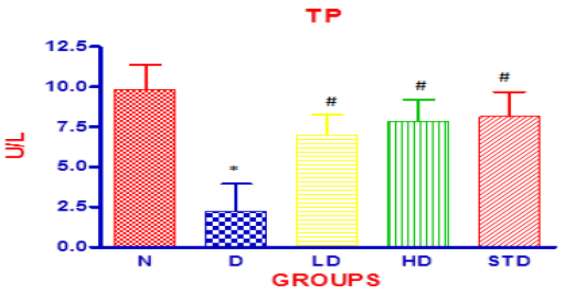

Fig. 7: Effect of phytol on TP. Values are expressed as MEAN±SEM. n=6.

*p	<0.05
** p	<0.01
*** p	p<0.001
#p	<0.05
##p	<0.01
### p	p<0.001

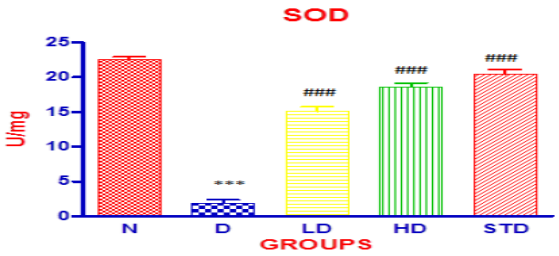

Fig. 8: Effect of phytol on SOD. Values are expressed as MEAN±SEM. n=6.

*p	<0.05
** p	<0.01
*** p	p<0.001
#p	<0.05
##p	<0.01
### p	p<0.001

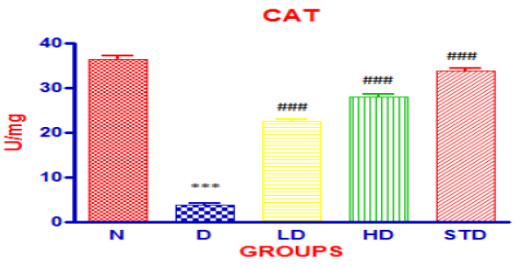

Fig. 9: Effect of phytol on CAT. Values are expressed as MEAN±SEM. n=6.

*p	<0.05
** p	<0.01
*** p	p<0.001
#p	<0.05
##p	<0.01
### p	p<0.001

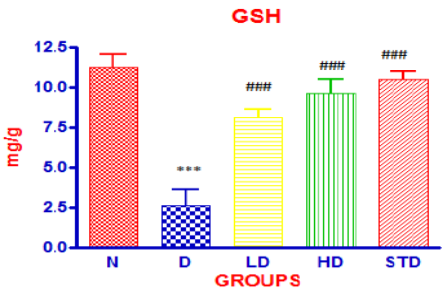

Fig.10: Effect of phytol on GSH. Values are expressed as MEAN±SEM. n=6.

*p	<0.05
** p	<0.01
*** p	p<0.001
#p	<0.05
##p	<0.01
### p	p<0.001

Discussion

The present study was aimed to evaluate the hepatoprotective effect of phytol. The hepatoprotective

effect of phytochemical phytoconstituent phytol in rats was studied during hepatic damage induced by toxic substance ethanol. In ethanol induced liver injury, ethanol produces a battalion of dose related harmful effects in the liver. The majority of ethanol is metabolized in the liver. Individuals who abuse alcohol by routinely drinking ethanol everyday are at risk for developing alcoholic liver disease. In chronics proteins, lipids and water accumulate in the liver cells. This causes enlargement of liver cells. Liver injury gives rise to lipid peroxidation which gives rise to free radical mediated liver cell injury. Also liver injury causes generation of reactive oxygen species and oxidative stress.[7] Phytol statistically reduced increased levels of SGPT, SGOT, ALP, TG, cholesterol and bilirubin in diseased animals. Phytol statistically increased reduced levels of TP, SOD, CAT and GSH in diseased animals (Fig. 1-10). The possible mechanism behind the hepatoprotection of phytol was might be due to its antioxidant activity, free radical scavenging activity, synergistic effect etc. In all, experimental model of the present study, the phytochemical phytoconstituent phytol showed potent hepatoprotective activity.

Conclusion

With the findings of the present study it can be concluded that both low (100 mg/kg) and high (200 mg/kg) doses of phytol showed statistically significant hepatoprotective activity in the ethanol induced pharmacological animal model. Phytol statistically reduced increased levels of SGPT, SGOT, ALP, TG, cholesterol and bilirubin in diseased animals. Phytol statistically increased reduced levels of TP, SOD, CAT and GSH in diseased animals.

Acknowledgement

The authors would like to thank Shree Devi Education Trust (R), Mangalore for providing the necessary facilities for carrying out the research work.

Conflict of Interest: None.

Reference

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How to cite this article: Gupta K, Taj T, Thansiya B, Kamath JV, Pre-clinical evaluation of hepatoprotective activity of phytol in wistar albino rats. *Int J Comprehensive Adv Pharmacol* 2019;4(1):17-20