JETIR.ORG ISSN: 2349-5162 | ESTD Year : 2014 | Monthly Issue
JOURNAL OF EMERGING TECHNOLOGIES AND INNOVATIVE RESEARCH (JETIR)
An International Scholarly Open Access, Peer-reviewed, Refereed Journal

EVALUATION OF HEPATOPROTECTIVE EFFECT OF ETHANOLIC EXTRACT OF *Arachis hypogaea* SEEDS

Jesny.s*[1], Aiswarya S Nair [2], Anusree s [3] Kiran K J[4] Prasobh G R[5]

[1] Mpharm Pharmacology student,Sreekrishna, college of pharmacy and research centre, Parassala,Thiruvanthapuram

[2] Mpharm Pharmacology student,Sreekrishna, college of pharmacy and research centre, Parassala,Thiruvanthapuram

[3]Associate professor,department of Pharmacology,Sreekrishna, college of pharmacy and research centre, Parassala,Thiruvanthapuram

[4] HOD,department of Pharmacology,Sreekrishna, college of pharmacy and research centre, Parassala,Thiruvanthapuram

[5] Principal, Sreekrishna, college of pharmacy and research centre,Parassala,Thiruvanthapuram

ABSTRACT

Traditional medicinal plants offer potential therapeutic benefits for liver health. On the basis of these considerations, the purpose of the present study was to evaluate the hepatoprotective activity of ethanolic extracts of Arachis hypogaea seeds. The hepatoptoprotective effect of EEAH was evaluated by various biochemical parameters for liver function like SGPT, SGOT, ALP, total bilirubin.Body weight measurements, liver weight analysis were also analysed .Histopathological examination was performed to evaluate liver tissue damage. The ethanol extract of Arachis hypogaea exhibits notable hepatoprotective effects, particularly at higher doses. The study suggest that EEAH has hepatoprotective activity, which may be due to the presence of resveratrol and betasetosterol which is associated with decrease in liver biochemical parameters and reduce hepatotoxicity.

Keywords: Arachis hypogaea, Beta setosterol, Hepatoprotective, Liver enzymes, Phytochemicals, Molecular docking, Resveratrol, Silymarin.

INTRODUCTION

There are several drugs which are plant derived and act directly or indirectly to protect the liver. . Due to the increasing prevalence of herbal medication in the management of liver disorders, liver herbal therapies are on therise in the recent time. Such hepatoprotective herbal agents are phenols, coumarins, lignans, essential oils, monoterpenes, glycosides, carotenoids, flavonoids, organic acids, lipids, alkaloids and xanthone derivatives.Protective elements

have the capacity to defend human and laboratory animals from the cytotoxicity caused by free radicals over production and many of these are antioxidants. Even some common vitamins, spices, and vegetables such as Vitamin E, turmeric, can cause this effect. In modern medicine, there is no fully effective and safe liver-protective drug. However Medicinal plants are frequently utilized for conditions like hepatitis and cirrhosis, especially in developing nations where they are valued for their minimal side effects. Traditional medicine, which relies heavily on plant materials, is used by about 80% of the global population. India, known as the "Medicinal Garden of the World," has a rich history of using herbal-based treatments for liver disorders.These traditional remedies are seen as effective alternatives to chemical-based drugs. Recently, phytoconstituents, the active compounds in plants, have gained attention for their hepatoprotective properties. These natural compounds have been used for a long time to treat liver disorders and are nowrecognized for their antioxidant capabilities, which help mitigate the harmful effects of free radicals. Arachis hypogaea, a kind of legume grown mainly for its seeds which might be consumable, is occasionally called the groundnut or peanut.[1-3]

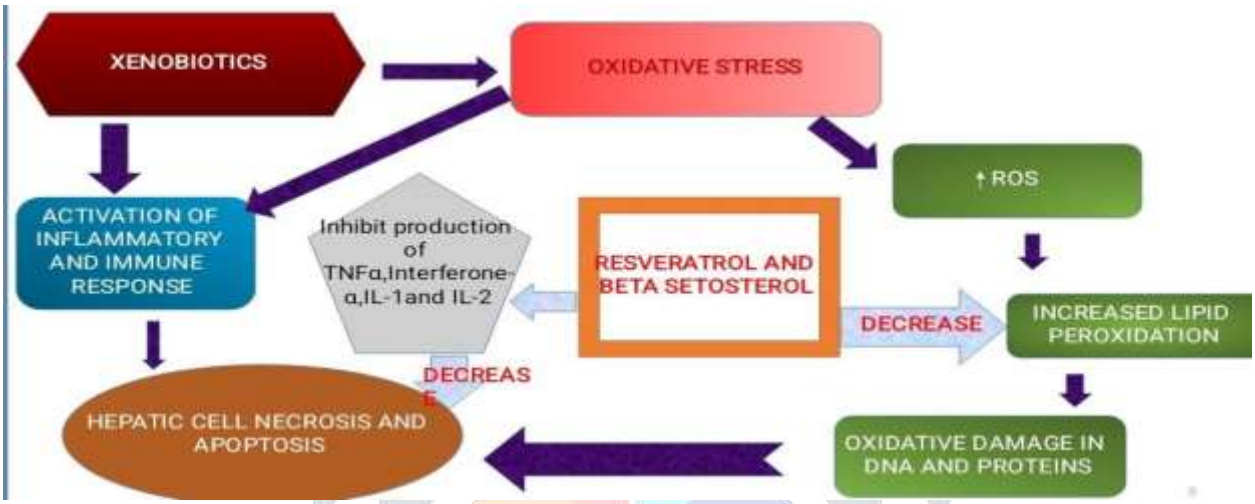


Fig no :1 mechanism of compound in arachis hypogaea seeds

MATERIALS AND METHODS

Arachis hypogaea seeds,wistar albino rats,paracetamol, silymarin was purchased

Methods:plant metirials was collected and authenticated. This was extracted by cold maceration procedure using absolute ethanol .To identify the different plant ingredients, a phytochemical screening was performed on the plant extract. Then total phenol and sterol content determined.

In silico computer simulations and docking studies are highly effective tools for examining the complementarity and interaction levels at the molecular scale between natural or synthetic compounds and their potential targets. The binding affinities of the selected compounds was done using the AutodockPyRxVina software. The chemical composition of these compounds were sourced from PubChem. Docking procedures were conducted with AutodockPyRxVina, and visualizations were created using PyMol software.

Male six to eight weeks old Wistar albino rattus, weight about 150-200g, were collected. The animals had been housed in a managed laboratory environment with 12 midnights and light, a temperature of 22±2ºC, and 45–60% humidity. Animals had been given seven days to acclimate.Acute toxicity study was done according to CPCSEA guideline 423.In the course of an acute toxicity investigation, the animals received an oral dose of 2000 mg/kg of EEAH and were analysed daily for period of 14 days. Notable changes are observed.[4-9]

PARACETAMOL INDUCED LIVER TOXICITY MODEL: EXPERIMENTAL DESIGNING

 Animals : Male Wistar albino rats

 Age and Weight : Six to eight weeks old, 150-200g.

 Number of animal needed : 30 Nos.

 Number of groups : 5 groups

 Animals in each group : 6 animal

Wistar albino rat will be used as experimental model.30 Animals are divided into 5 groups. GroupI (normal control) animals acquired normal saline (1ml/kg p.o). Group II(disease control) animal received normal saline and paracetamol (2g/kg p.o).Group III animal received silymarin(100mg/kg) which serves as standard. Group IV and V test groups received 200 and 400 mg/kg EEAH. Hepatotoxicty induction was done on 10th day. All group except control group animals received paracetamol 2g/kg orally at a single dose. Animals will be given ketamine and xylazine for anesthesia 24 hours after the hepatotoxin is administered, and 1 milliliter of blood will be drawn from the retroorbital. various biochemical estimation SGPT, SGOT, ALT, TB was done. Histopathology estimation was also done.

STATISTICAL ANALYSIS: The most recent version of Graph Pad Prism was utilized for statistic analysis and interpretation. The data is presented as mean ± SEM, with n = 6. Students' "t" test was used to compare the group , and one way ANOVA was used for the multiple instance, which was then followed by the Dunnetts test.[10,11]

RESULT AND DISCUSSION

The study involved a detailed preparation and evaluation of Arachis hypogaea seed extract. The seeds were collected from Thiruvananthapuram in the month of January. After collecting it was authenticated from department of Botany, Nesamony memorial Christian college, marthandam TamilNadu. Later the samples, were thoroughly washed with fresh water and water content was removed by drying. After that peels of the seeds are removed and it is pulverized into a coarse powder and subjected to ethanol extraction via cold maceration. Folowing phytochemical estimation, alkaloids, and steroids was confirmed in the extract. There were 128.8µg/mg of sterol and 38.9µg/mg of total phenolic content.the existance of phenolicsubstances,flavonoids,tannins,terpenoids

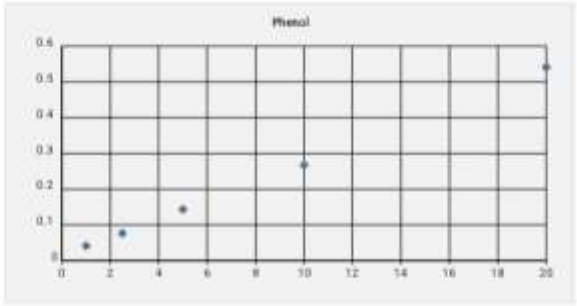
● Percentage yield of seeds extract of *Arachis hypogaea.*

Plant name	Part used	Method of extraction	Solvent used	Percentage yield(%w/w)
Arachis hypogaea	seeds	maceration	Ethanol	21%W/W

Table no: 1 percentage yield of extract

DETERMINATION OF TOTAL PHENOL CONTENT

Standards	Concentration of gallic acid (µg/ml)	OD at 750 nm
S1	1	0.041
S2	2.5	0.076
S3	5	0.143
S4	10	0.267
S5	20	1.242

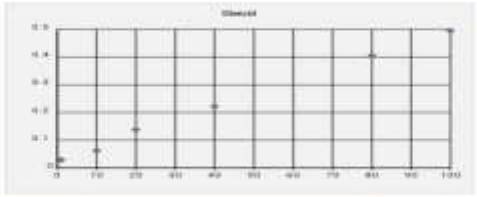


Sl. No.	OD at 750 nm	Concentration of phenol in Gallic acid
1	0.113	38.9

Table no:2 total phenol content

DETERMINATION OF TOTAL STEROID CONTENT

Standards	Concentration Cholesterol (µg/ml)	OD at 540 nm
S1	1	0.029
S2	10	0.062
S3	20	0.138
S4	40	0.221
S5	80	0.405
S6	100	0.495



Sample code	OD at 540nm	Concentration of steroid (µg/mg)
AH	0.034	128.8

Table no:3 total sterol content

Molecular docking has become an increasingly valuable tool in drug discovery, allowing for the assessment of how potential therapeutic agents interact with specific biological targets. In this study, molecular docking was employed to explore the interactions of resveratrol (RES) and beta-sitosterol(BSS) with three hepatic receptors: 3ETR, 5WB1, and 2YCW. These receptors are implicated in liver function and damage response.The docking studies revealed that RES and BSS exhibit strong binding affinities with the hepatic receptors, particularly with receptor 3ETR. This suggests that both compounds have significant potential for hepatoprotection. Both RES and BSS demonstrated hepatoprotective activity through their interaction with hepatic receptors and their capacity to combat oxidative stress and inflammation. These findings underscore the therapeutic potential of these compounds in liver-related disorder .

HYDROGEN BOND INTERACTION AND BINDING AFFINITY OF RESVERATROL AND BETASETOSTEROL WITH RECEPTORS

RECEPTOR	NUMBER OF HYDROGEN BOND INTERACTIONS		BINDING AFFINITY (kcal/mol)	
	RES	BSS	RES	BSS
3ETR	5	6	-8.6	-8.8
5WB1	4	5	-8.2	-8.4
2YCW	3	2	-7.9	-6.9

Table no:4 hydrogen bond interaction and binding affinity of compounds with receptors

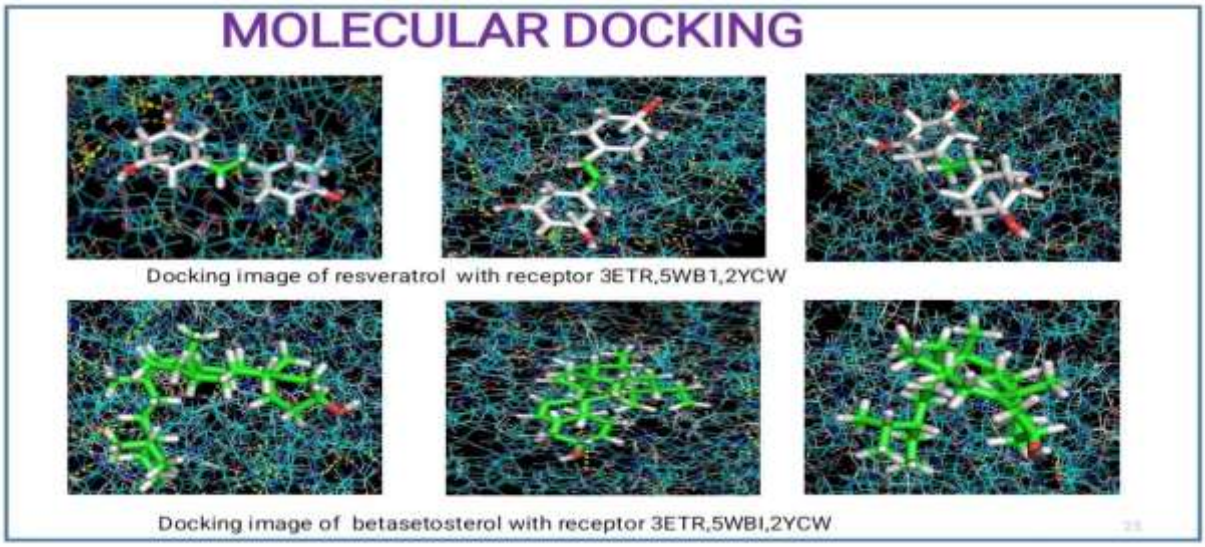


Fig no :2 docking images

The acute toxicity study of EEAH was conducted to in order for assessing the safety characteristics of the extract, following the OECD-423 guidelines. The study utilized a dose of 2000mg/kg, which is a high dose intended to evaluate the potential for acute toxic effects.The results from this study indicated that EEAH possesses a substantial safety margin. Specifically, no fatalities were observed at the 2000 mg/kg dose, and there were not remarkable alterations in the rats autonomic functions or general behaviour. This lack of adverse effects confirms the extract's safety at this doselevel. So 200mg/kg and 400 mg/kg doses were selected as low and high dose for the study respectivelyThe hepatoprotective effects of the EEAH were evaluated using Wistar albino rats. Model of paracetamol-induced injury to the liver are selected for the study. This method are selected based on the reference of the article by Muhammad Saidurrahman et al. The present study involved five experimental groups, each consisting of six animals. The groups included a control group which administered with normal saline of 1ml/kg for 10 days, a disease group receive normal saline for 9 days and paracetamol on 10th day,a standard group administered with silymarin 100mg/kg for 9 days and paracetamol 2g/kg on 10th day and two treatment groups: receiving different doses of the Arachis hypogaea extract (200mg/kg and 400 mg/kg) for 9 days and paracetamol 2g/kg on 10th day. On 11th day animals will be sacrificed and blood and liver

collected for further examinations.The animals' beginning and ending body weights have been collected for each group; the information is shown in Table .According to the statistics, the control group's body weight increased during the course of the trial although they simply received water and a conventional diet. On the other hand, the illness group, which received a hepatotoxin, showed a notable decrease in their ultimate body weight.This is because paracetamol causes hepatotoxicity. When compared to the disease group, treatment with the conventional medicine led to a significant rise in body weight, indicating a preventive benefit against the weight loss commonly associated with liver impairment. In the same way, the AHE-treated groups' body weight increased in comparison to the illness group. This improvement suggests that the extract aids in body weight restoration and lessens the degree of hepatotoxicity.

The relative liver weights of the various groups in this study, as detailed in Table 5.8, animals in the disease group displayed an increased relative liver weight correlate to the control group. This increase in liver weight is indicative of hepatic inflammation or damage rather than growth. The group treated with standard showed a decraese in liver weight relative to the disease group, bringing it closer to control levels. This reduction suggests that silymarin effectively counteracts liver damage and inflammation. The treatment groups receiving the Arachis hypogaea extract also demonstrated reduced liver weights compared to the disease group. In summary, the ethanol extract of Arachis hypogaea demonstrated a notable hepatoprotective effect in the experimental model, with both tested doses improving body weight and reducing liver weight relative to the disease group. However, silymarin, the standard hepatoprotective drug, displayed greater effectiveness in this study.

e

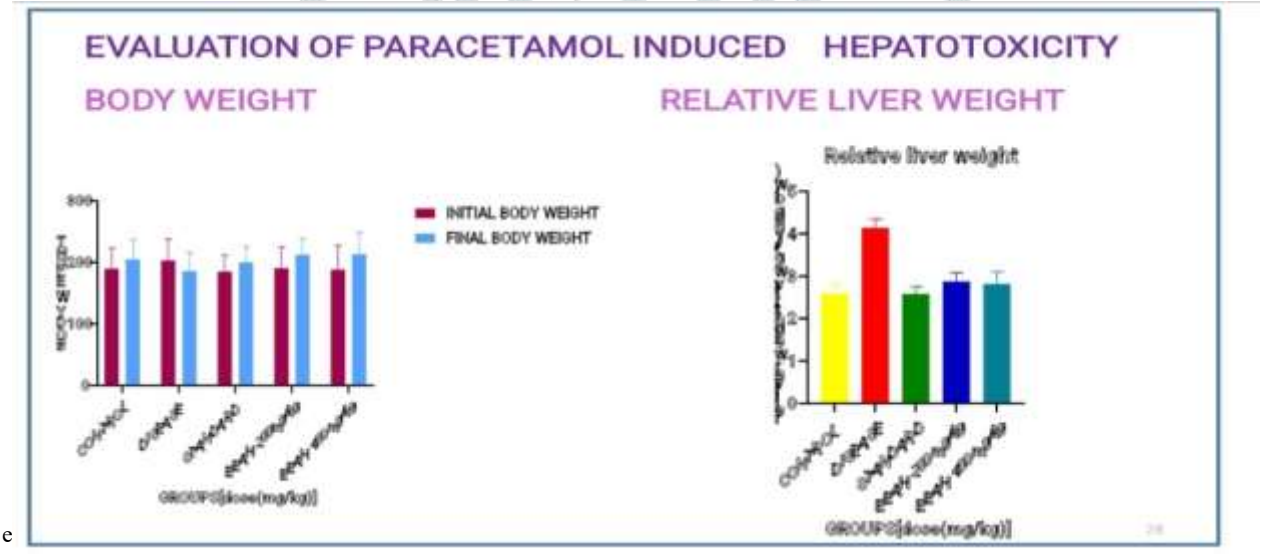


Fig no 3: estimation of body weight and liver weight

To compare the ethanol extract of Arachis hypogaea with the conventional hepatoprotective medicinal drug and examine the hepatoprotective traits of the extract a biochemical study involving liver function markers was carried out. For each group, the findings were presented as mean ± SEM, with n =6. The statistical evaluations were carried out using GraphPad Prism, employing Student's t-test forcomparisons among pairs and one-way ANOVA accompanied by Dunnett's test for multiple analyses.

In this study assessing liver protection, paracetamol administration led To greater levels of SGPT, SGOT, total bilirubin and ALP indicating liver damage. Treatment with Silymarin considerably lowered these increased enzyme concentrations. ($p < 0.0001$), highlighting its hepatoprotective properties. Both doses of EEAH also reduced SGPT, SGOT, total bilirubin and ALP levels compared to the disease group, indicating hepatoprotective effects. While EEAH showed promising results, silymarin was consistently more effective in reducing liver enzyme levels, suggesting it is a superior hepatoprotective agent. Despite this, the high dose of EEAH demonstrated significant liver protection,making it a therapeutic option for liver disorders. Additional investigation is required to examine the mechanisms. and clinical efficacy of EEAH. The obtained results are in agreement with the findings reported in these literature Fatma A. Elshibani et al in their paper, examined and validated A clinicall pavarii's antioxidant and hepatoprotective effects using a wide range of

biochemical and histological markers in a rat model of PAR-induced hepatotoxicity. The delivery of PAR resulted in immediate liver injury, as seen by a considerable increase in ALT, AST, and TB levels, according to liver function markers. On the other hand, pretreatment with ARB extract significantly decreased AST,ALT and TB, which mitigated PAR-induced hepatotoxicity.

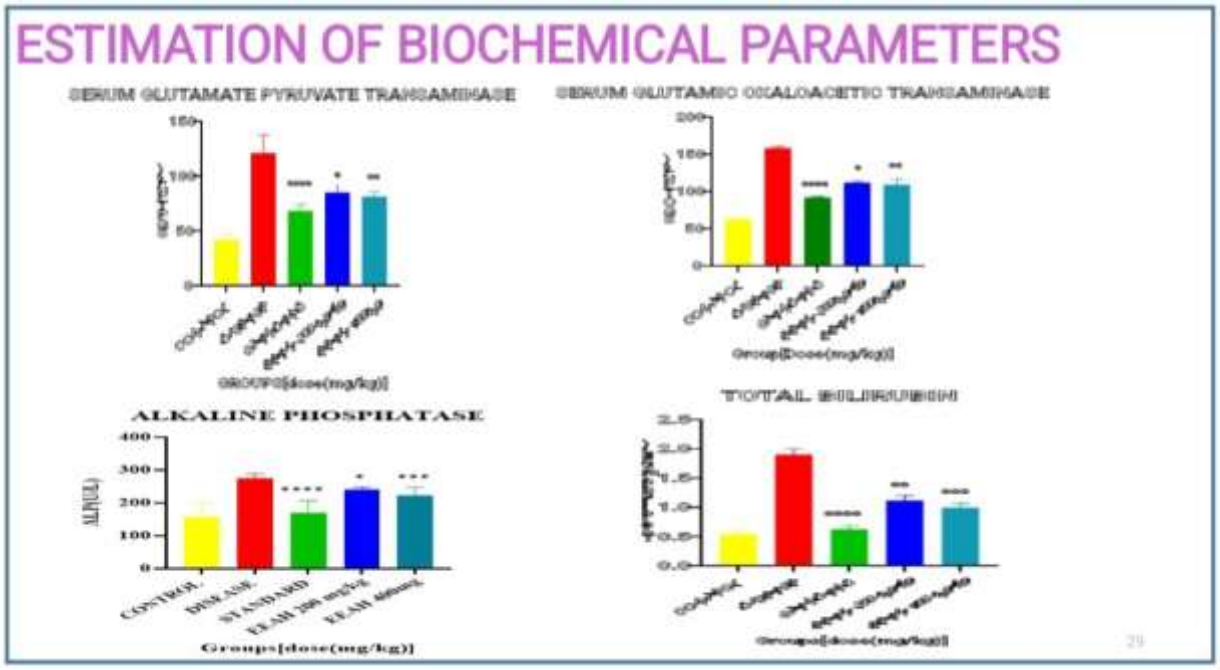


Fig no:4 estimation of biochemical parameters

Histopathological examination of liver tissues revealed distinct patterns across different groups.In the control group, liver architecture was normal with well-defined lobular structures and regular hepatocyte arrangement. The paracetamol-treated disease group displayed significant liver damage, including disrupted architecture, hepatocyte degeneration, and increased intracellular spaces. Silymarin-treated rats showed mild inflammatory changes and sinusoidal congestion, but also signs of hepatocyte regeneration, indicating its hepatoprotective effect. The low-dose EEAH group exhibited mild sinusoidalcongestion and degenerative changes, with some hepatocyte regeneration. The high-dose EEAH group demonstrated near-normal liver architecture with improved hepatocyte arrangement and less pronounced congestion, indicating strong hepatoprotective activity of Arachis hypogaea.

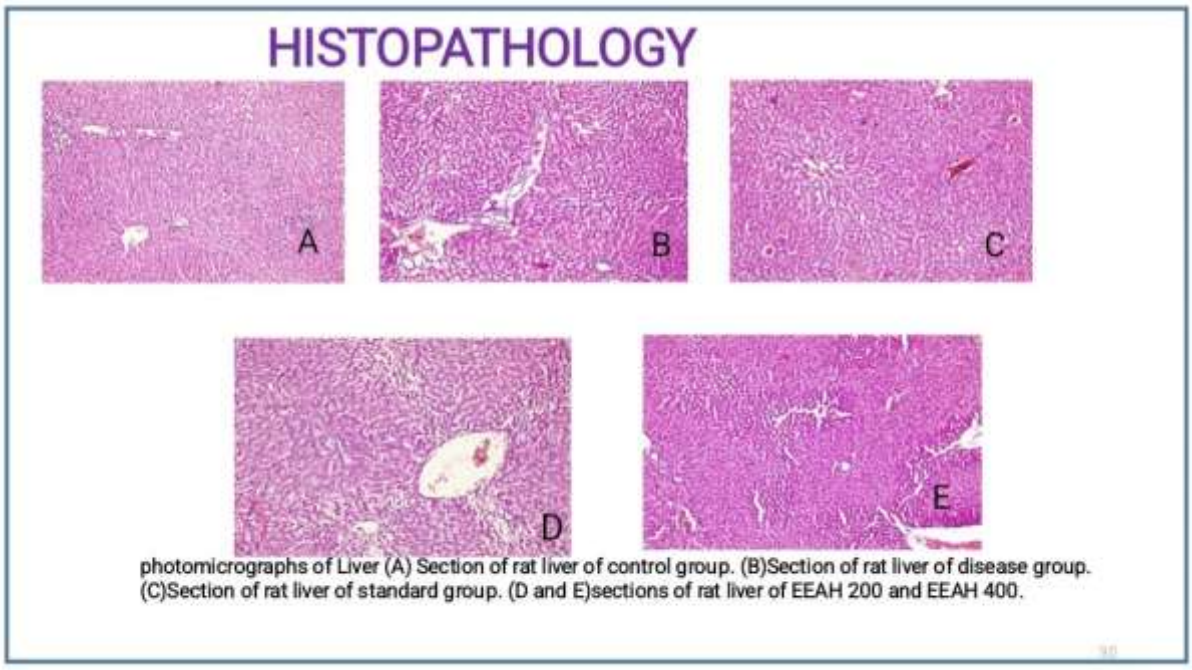


Fig no :5 histopathology of liver

The study demonstrates that the ethanol extract of Arachis hypogaea (EEAH) exhibits significant hepatoprotective effects, particularly at higher doses. The extract's efficacy is supported by improvements in body weight, reductions in liver weight, and favorable changes in liver enzyme levels(SGPT, SGOT, and ALP). Histopathological evaluations further confirm that EEAH promotes liver cell regeneration and reduces hepatic damage. Arachis hypogaea's hepatoprotective properties are attributed to the bioactive substances BSS and RES. By scavenging reactive oxygen species, these chemicals minimize oxidative stress, lower lipid peroxidation, and stop the formation of inflammatory mediators. This is how they exercise their hepatoprotective action. These results imply that EEAH may be a useful therapeutic agent for liver diseases;nevertheless, more investigation is required to completely comprehend its mechanisms and therapeutic possibilities.

CONCLUSION

The ethanol extract of Arachis hypogaea (peanut) seeds was assessed for its hepatoprotective Effects. Botonical compounds study indicated the existence of significant bioactive compounds, Including phenolic compounds, flavonoids, and phytosterols. These compounds are likely in charge for The observed hepatocellular defence efficacy. Specifically, resveratrol and beta-sitosterol were identified As key components, showing strong binding affinities with hepatic receptors implicated in liver damage.Acute toxicity study reveled that the EEAH are safe within a 2000 mg/kg dose. The extract Demonstrated notable hepatoprotection in Wistar rats, with improvements in body weight, reductions in Liver weight, and a lower degree of liver enzymes (SGPT, SGOT, and ALP). Histological analysisFurther verified the restoration of liver architecture, with the high-dose group exhibiting near-normal Liver tissue structure.

While Arachis hypogaea extract shows promising potential as a therapeutic agent for liver Disorders, silymarin, a standard hepatoprotective agent, proved to be more effective in mitigating liver Damage. This study highlights the extract's capacity to lessen oxidative stress and inflammation in the Liver. Future research should focus on isolating specific bioactive compounds from Arachis hypogaea And elucidating their mechanisms of action.

REFERENCE

1. C. Delgado et al,models of hepatoprotective activity assessment,medicina universitaria.2015;17(69):222-228
2. Harshad Devarbhavi et al, Global burden of liver disease: 2023 update,journal of hepatology.2023;79:516-537
3. Renata Miranda et al,Chemical composition and biological activities of Arachis species,journal of agricultural and food chemistry.2011;59:4321-4330
4. Pilin francis et al ,Drug induced hepatotoxicity,national library of medicine,2022;
5. Anton et al,Silymarin as supportive treatment in liver disease,adv ther, 2020;37:1279-1301
6. Supat et al, hepatoprotective and therapeutic effect of resveratrol;fundam clin pharmacology,2021;1-9
7. Gabriel casimiro et al Friable Calluses of a Brazilian Peanut Cultivar Increased Cytotoxic Activity against K562 Human Leukaemia Cells, *Brazilian Archives of Biology and Technology*. 2023;66
8. Bogdan et al,Cardioprotective and Hepatoprotective Potential of Silymarin inParacetamol-Induced Oxidative Stress, Pharmaceutics. 2024;16: 520
9. Mahboubeh Mansourian *et al* Hepa,toprotective and antioxidant activity of hydroalcoholic extract of Stachys pilifera. Benth on acetaminophen-induced liver toxicity in male rats,heliyon,2020;e03029
10. A Lakshmana Rao et al Evaluation of hepatoprotective activity of indigofero barberi in rats against Paracetamol induced hepatic injury,adv Inv Pha The Medic1:1-9,2018
11. Mohammad Saidurrahman et al,Evaluation of hepatoprotective activity of ethanolic extract od pterocarpus marsupium Roxb. Leaves against paracetamol-induced liver damage via reduction of oxidative stress,phytomedicine plus,2022;10311