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# HEPATOPROTECTIVE EFFECT OF *ABELMOSCHUS FICULNEUS* ROOT EXTRACT AGAINST DRUG INDUCED HEPATOTOXICITY CAUSED BY THE COMBINED ACTION OF ISONIAZID AND RIFAMPICIN

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## ABSTRACT

The present study was focused on the hepatoprotective effect of *Abelmoschus ficulneus* root extract against drug induced hepatotoxicity caused by the combined action of Isoniazid and Rifampicin in rats. Administration of INH and RIF (each at 50 mg/kg, i.p.) for 15 days induces the hepatotoxicity by altering the levels of hepatic marker enzymes, lipid profile parameters, lipid peroxides, oxidative stress markers and histopathological parameters. Treatment with methanolic extract of *Abelmoschus ficulneus* at 100, 200 and 400 mg/kg, p.o significantly ameliorated all altered parameters along with histopathological findings. The results revealed that *Abelmoschus ficulneus* root extract has exhibited more significant protection from drug induced hepatotoxicity at 400 mg/kg and it was well comparable to the standard drug silymarin at 100 mg/kg.

**Keywords:** Hepatotoxicity, Hepatoprotective effect, *Abelmoschus ficulneus*, Lipid peroxidation, Liver enzymes.

## 1. INTRODUCTION

Liver is the primary site for metabolism of almost all drugs because it possesses a large variety of enzymes with large amounts. These enzymes are versatile and non specific in metabolizing a large number of drugs hence damage of this organ is common, leads to toxicity called drug induced hepatotoxicity [1].

Drug induced hepatotoxicity is the most common, effective and frequent cause because liver is the main target of drugs for concentrating, metabolizing. This toxicity is hard to determine because of difficulties in detection, diagnosis and lack of exposure and the death rate is high up to 10%. Drugs can cause liver toxicity in many ways, some drugs increases drug metabolizing ability of enzymes, some dugs cause dose dependent, idiosyncratic toxicity etc [2].

Drugs and its metabolites are an important cause of liver injury, more than 900 drugs and their toxins have been reported to cause liver injury and it is the main reason for a drug to be withdrawn from the market [3]. In India, more than 50 different agents or classes of drugs are associated with liver injury. In these agents, antitubercular drugs is the primary class (46.4%) of drug induced liver toxicity followed by complementary and alternative medicines (13.9%), antiepileptic drugs (8.1%), antibiotics (6.5%), antimetabolites (3.8%), antiretroviral drugs (3.5%), NSAID (2.6%), hormones (2.5%), statins (1.4%) and others [4].

Generally antitubercular drugs used to cure tuberculosis, are the most common and effective drugs to induce hepatotoxicity. Tuberculosis is the serious bacterial infectious disease mostly affects the lungs. Approximately, one third of the world population is affected by tuberculosis and particularly observed in developing countries [5]. Every year 10 million people are infecting with tuberculosis worldwide. Even though being a preventable and treatable disease, 1.5 million people die each year. In India, more than one million people are affecting per year [6]. In the treatment of tuberculosis, WHO recommends 2 months intensive treatment phase followed by four months continuation phase. Generally in tuberculosis treatment, drugs are always used together to prevent drug resistance. Isoniazid (INH) and rifampicin (RIF) are the first line drugs used in combination and highly effective in the treatment of TB [7]. During the treatment of TB, serious and sometimes fatal liver problems may occur with INH and RIF. Analysis of studies involves that risk

of liver problems is highest in adults between the ages of 35 to 65 and shown the liver toxicity up to 2.6% [8]. So liver function need to be checked every month in the treatment of TB with INH and RIF.

Hepatotoxicity with INH and RIF is associated with its metabolism. These drugs are metabolized by various hepatic enzymes of cytochrome P450 family. During the metabolism, INH generates toxic metabolites by the hydrolytic pathway such as mono acetyl hydrazine, hydrazine and its related compounds and also it inhibits cytochrome P450 1A2 reductase, an enzyme involved in the detoxification of toxic metabolites. Rifampicin is a potent inducer of various isoforms of cytochrome P450 enzyme, in presence of RIF, INH metabolism increases ten-fold by hydrolytic pathway leads to further increase in the concentration of INH toxic metabolites. This reactive metabolite formation leads to hepatocellular necrosis, mononuclear cell infiltration, oxidative damage and increased lipid peroxidation resulting from reactive oxygen species produced due to an imbalance of antioxidant mechanism [9].

Unfortunately, no single drug is available for preventing the hepatotoxicity of INH and RIF. So the usage of herbal medicines has been increasing worldwide, due to its harmlessness, lesser side effects and easy availability. In India, herbal products are used as traditional medicine for the treatment of liver complications because these consist of phytochemicals which acts as good hepatoprotective agents which can cure the damage of the liver with INH and RIF by many routes mainly acting on enzymes of cytochrome P450, inhibit the microsomal drug metabolizing enzymes and acts as an antioxidant on free radicals there by reduces the oxidative stress and oxidative damage observed in hepatotoxicity [10].

*Abelmoschus ficulneus*(L.)Wight & Arn. (family: Malvaceae) commonly known as White wild musk mallow found in wastelands and cultivated fields in most districts of India and also other countries. It is a shrub, 2 to 5 feet tall and 2 to 6 feet across, flowers are about an inch in diameter, either creamish or white with a rose center, leaves are palmate 5 to 8 cm long and 4 to 7 cm wide, with a circular shape. Leaves are rough on both sides and have 3 to 5 lobes. Capsules are oblong-ovoid, 5 angled, shortly beaked, tomentose, seeds are globose, sulcate, slightly pilose [11, 12]. It has been used as traditional herbal medicine in asthma, spasm, varicose veins, disorders of spleen, pectorial lesions, inflammation, stress, insect bites, scorpion and snake bites, fever, stomachic etc. Traditionally ground seeds are used in asthma, leaves are used in constipation, roots helps in overcoming deficiency of calcium in the human body and acts as a good heart tonic, its paste helps in healing cuts and bruises. Crushed roots with water treats jaundice and other GIT problems [13].

Phytochemically, leaves have beta-sitosterol and beta-D glucoside, flowers have anthocyanins. Petals have beta-sitosterol, flavonoids. Seeds have an essential oil [14]. Antimicrobial activity was evaluated from the leaf extract [15]. Antioxidant activity and neutraceutical composition has been reported from the fruit extracts [16]. Four Lignans along with myriceric acid were isolated from methanolic extract of *Abelmoschus ficulneus* stem bark [17]. Amino acid and fatty acid composition was estimated from the seed oil of *Abelmoschus ficulneus* [18].

Hepatoprotective activity of *Abelmoschus ficulneus* roots have not been reported earlier, so the present study was designed to evaluate the hepatoprotective effect of *Abelmoschus ficulneus* against drug induced hepatotoxicity caused by the combined action of Isoniazid and Rifampicin.

## 2. MATERIAL AND METHODS

### 2.1. Drugs and chemicals

Standard Isoniazid and rifampicin drugs were purchased from Sigma-Aldrich, India. Silymarin was procured from Sigma-Aldrich, China. Biochemical kits for estimation of AST, ALT, ALP, ALB, TP, TB, DB, LDH were purchased from Merck specialities Pvt. Ltd, Mumbai, India. TCA and TBA were purchased from Himedia, Mumbai, India and all the used chemicals and reagents were of analytical grade.

### 2.2. Collection and extraction of plant material

The roots of *Abelmoschus ficulneus* were collected from the wastelands in Gangapuram village, Yadadri district, Telangana, India and the plant material was authenticated by Prof. V.S. Raju (Botanist) Department of Botany, Kakatiya University, Warangal. A voucher specimen of the plant having number KU/UCPSC/53 was kept in the herbarium of Department of Pharmacognosy and Phytochemistry in University college of Pharmaceutical sciences, Kakatiya University, Warangal. One kilogram of fresh roots was taken and washed under running tap water, shade dried and coarsely powdered. This powdered plant material was extracted with methanol by maceration technique for seven days and filtered. The filtrate was concentrated to dryness by using rotary evaporator and percentage yield

was calculated (5.6%) then stored in desiccator. The obtained methanolic extract was tested for various phytoconstituents like alkaloids, glycosides, flavonoids, tannins, terpenoids, saponins etc. by using different chemical tests.

#### *2.2.1. Estimation of total phenolic content*

The total phenolic content in methanolic extract of *Abelmoschus ficulneus* (AFME) was estimated by using Folin-Ciocalteu colorimetric method using gallic acid as a standard and the amount of total phenolics was expressed in terms of gallic acid equivalent (GAE) [19].

#### *2.2.2. Estimation of total flavonoid content*

The total flavonoid content in AFME was estimated by using Aluminium chloride colorimetric method using rutin as a standard and the amount of total flavonoids was expressed in terms of rutin equivalent (RE) [19].

### 2.3. Experimental animals

Both Male and Female Wistar rats weighing 150-200 grams were purchased from Vyas Labs, Hyderabad, India. They were kept in polypropylene cages and housed for acclimatization at 22±3°C with a 12 hour light/dark cycle for one week prior to the experiment with permission from institutional animal ethical committe (IAEC/12/UCPSC/KU/2020) Rats were fed with standard pelleted diet, drinking tap water *ad libitum*.

### 2.4. Acute toxicity study

Acute toxicity study was performed on the methanolic extract of plant according to the OECD-423 guidelines [20]. Female Wistar rats were used in this study. The animals were fasted overnight with only water accessible before administration of test dose. All the animals were observed individually after dosing, during the first 24hours and then daily for 14 days to observe the mortality and signs of toxicity.

### 2.5. Experimental design

Wistar male rats were randomly divided in to six groups with six animals in each group (n=36). Treatment of each group was as follows: Group I- normal control, animals of this group were administered normal saline orally once daily for 15 days. Group II- was toxic control and they were intoxicated with INH and RIF to induce hepatic damage (50 mg/kg, each by i.p) once daily for 15 days, Group III- Standard group, in which animals were treated with silymarin (100 mg/kg, p.o) once daily for 15 days an hour before administration of INH and RIF. Group IV, V and VI- Test (Treated) groups 1, 2 and 3, animals of which received methanolic extract of *A.ficulneus* (100, 200 and 400 mg/kg p.o) once daily for 15 days an hour before administration of INH and RIF. After administration of last dose, animals were allowed to be fasted overnight. On the next day, whole blood was withdrawn from the rats by sino-orbital puncture with the overdose of diethyl ether then the animals were sacrificed and liver was separated immediately and rinsed in cold saline, blotted, dried, weighed and used for preparation of liver homogenate, histopathological findings [21-23].

### 2.6. Biochemical parameters

The collected blood was allowed to coagulate at room temperature then centrifuged at 3000 rpm for 10min to separate the serum. The obtained serum was used for estimating the biochemical parameters like aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatise (ALP), total bilirubin (TB), direct bilirubin (DB), total protein (TP), albumin (ALB), lactate dehydrogenase (LDH) and lipid profile parameters [24] including HDL-cholesterol, LDL-cholesterol, Cholesterol, triglycerides content by using commercially available standard assay kits with autoanalyzer.

### 2.7. Oxidative stress parameters

A portion of collected liver tissue (10%) was homogenised with basic phosphate buffer (pH 7.4) by using tissue homogenizer and homogenate was centrifuged at 3000rpm for 15 min at 4°C. The obtained homogenate was used to estimate lipid peroxidation (LPO)[25], glutathione (GSH)[26], catalase (CAT)[27] and superoxide dismutase (SOD)[28] levels by using standard procedures.

### 2.8. Histopathological study of liver

The remaining portion of collected liver tissue was fixed in 10% buffered neutral formalin solution, embedded in paraffin, cut in to sections of 3-5µm and stained with hematoxylin-eosin. Finally, microscopic observation was done by using Digital Motic Microscope under 100X magnification.

### 2.9. Statistical analysis

The results of data obtained were analyzed by one-way analysis of variance (ANOVA) followed by Dunnett's method using graph pad prism 9.0 and all the results

were expressed as mean ± Standard deviation (SD). The value of $p<0.05$ was considered as statistically significant.

## 3. RESULTS

### 3.1. Phytochemical analysis

Preliminary phytochemical analysis revealed the presence of alkaloids, glycosides, steroids, flavonoids, saponins, terpenoids and phenolic compounds in methanolic extract of *Abelmoschus ficulneus* roots. The total phenolic content in extract was found to be 89.11±2.14 mg of GAE per gram of dry extract and the total flavonoid content in extract was found to be 25.75±1.55 mg of RE per gram of dry extract.

### 3.2. Acute toxicity study

Rats administered with AFME did not show any toxicity symptoms during the first 24hours and no mortality occurred until the period of 14 days with a dose level of up to 2000 mg/kg body weight.

### 3.3. Effect of AFME on body weight and liver weight

The effect of AFME on body weight and liver weight in INH and RIF induced hepatotoxicity in rats was shown in table 1. The final body weight of rats intoxicated with INH and RIF significantly decreased compared with normal control. Groups treated with AFME at 100, 200 and 400 mg/kg and standard drug silymarin at 100 mg/kg significantly increased the body weight compared with the toxic group. The Liver weight, relative liver weight of rats induced with INH and RIF increased significantly when compared with normal rats. Significantly decreased liver weight and relative liver weight was observed in the groups of rats treated with test doses, standard dose compared to toxic group of rats.

**Table 1: Effect of AFME on body weight and liver weight in INH and RIF induced hepatotoxicity in rats**

| Groups | Initial body weight(g) | Final body weight(g) | Liver weight(g) | Relative liver weight(%) |
|---|---|---|---|---|
| I-Normal | 175.5±7.56 | 192.3±10.5 | 6.55±0.12 | 3.40±0.12 |
| II -Toxic | 170.8±5.75 | 145.5±8.41[a] | 9.51±0.17[a] | 6.53±0.16[a] |
| III-Standard | 165.6±6.54 | 187.8±5.81[b] | 6.75±0.18[b] | 3.59±0.07[b] |
| IV-Test1 | 170.4±5.84 | 178.5±6.61[c] | 8.55±0.11[c] | 4.78±0.12[c] |
| V -Test2 | 178.7±8.12 | 197.5±8.22[b] | 7.86 ±0.11[b] | 3.97±0.20[b] |
| VI -Test3 | 168.7±6.33 | 195.8 ± 9.21[b] | 7.10±0.07[b] | 3.62±0.10[b] |

*Values are expressed as mean ± SD, n=6. [a]P<0.001 values-compared to normal control. [b]P<0.001, [c]P<0.01, [d]P<0.05 values-compared to toxic control.*

### 3.4. Effect of AFME on serum biochemical parameters

The effect of AFME on biochemical parameters of serum was shown in table 2. Induced hepatotoxicity was observed in INH and RIF treated group with significantly increased levels of AST, ALT, ALP, LDH, TB, DB and decreased TP, ALB levels were observed compared to normal group. In standard, extracts treated test groups the AST, ALT, ALP, LDH, TB, DB levels were decreased and TP, ALB levels were increased significantly when compared to toxicity induced group; it indicates the standard and test groups were protected from hepatotoxicity caused by INH and RIF. From the three test doses, AFME at 400 mg/kg was better protected and it was well comparable to that of standard drug silymarin at 100 mg/kg.

### 3.5. Effect of AFME on serum lipid profile

The effect of AFME on lipid profile of serum in INH and RIF induced hepatotoxicity was shown in table 3. Rats treated with only INH and RIF showed significant elevated levels of cholesterol, triglyceride, LDL-cholesterol and depleted levels of HDL-cholesterol when compared to normal control. Rats treated with extracts, standard along with INH and RIF showed significantly decreased levels of cholesterol, triglyceride, LDL-cholesterol and increased levels of HDL-cholesterol as compared to toxic control indicates that rats treated with standard and test doses showed better protection from toxicity without any altered lipid profile parameters.

### 3.6. Effect of AFME on oxidative stress parameters

The effect of AFME on oxidative stress parameters of liver tissue protein in INH and RIF induced hepatotoxicity was shown in table 4. The hepatic CAT, GSH, SOD levels were significantly decreased and hepatic LPO levels in tissue homogenate were increased in toxic group II compared to normal group I. When

treated with standard dose in group III, test doses in group IV, V, VI along with INH and RIF the activities of CAT, GSH, SOD were significantly enhanced and the activity of LPO was significantly reduced as compared to toxic alone in group II.

**Table 2: Effect of AFME on biochemical parameters of serum in INH and RIF induced hepatotoxicity**

| Groups | AST(U/L) | ALT(U/L) | ALP(U/L) | LDH(U/L) | TB(mg/dl) | DB(mg/dl) | TP(g/dl) | ALB(g/dl) |
|---|---|---|---|---|---|---|---|---|
| I-Normal | 48.58±3.51 | 54.17±2.53 | 150.91±5.42 | 180.17±4.24 | 0.51±0.03 | 0.25±0.02 | 6.3±1.21 | 3.9±0.54 |
| II -Toxic | 85.64±4.27[a] | 119.19±8.55[a] | 259.13±9.11[a] | 495.13±9.54[a] | 2.63±0.51[a] | 1.89±0.55[a] | 2.1±0.32[a] | 2.2±0.04[a] |
| III-Standard | 54.13±2.78[b] | 59.13±3.11[b] | 145.24±6.52[b] | 160.21±7.51[b] | 0.55±0.03[b] | 0.27±0.02[b] | 5.8±1.11[b] | 3.7±0.25[b] |
| IV-Test1 | 73.51±3.55[c] | 94.15±6.37[c] | 203.51±9.25[c] | 350.31±9.95[c] | 1.13±0.10[c] | 0.81±0.08[c] | 3.7±0.31[c] | 2.5±0.24[c] |
| V -Test2 | 67.87±3.45[b] | 83.58±5.55[b] | 159.54±5.69[b] | 239.43±9.56[b] | 0.82±0.05[b] | 0.43±0.08[b] | 4.6±0.27[b] | 3.1±0.33[b] |
| VI -Test3 | 59.35±2.55[b] | 65.31±4.58[b] | 139.42±4.54[b] | 189.55±5.63[b] | 0.63±0.02[b] | 0.29±0.04[b] | 5.4±0.51[b] | 3.5±0.27[b] |

*Values are expressed as mean ± SD, n=6. [a]P<0.001 values-compared to normal control. [b]P<0.001, [c]P<0.01, [d]P<0.05 values-compared to toxic control.*

**Table 3: Effect of AFME on lipid profile of serum in INH and RIF induced hepatotoxicity**

| Groups | Cholesterol (mg/dl) | Triglycerides (mg/dl) | HDL-cholesterol (mg/dl) | LDL-cholesterol (mg/dl) |
|---|---|---|---|---|
| I-Normal | 65.24±2.55 | 53.66±2.17 | 39.55±1.20 | 26.35±0.84 |
| II -Toxic | 111.54±5.12[a] | 125.45±5.54[a] | 19.53±0.71[a] | 56.87±6.55[a] |
| III-Standard | 68.56± 3.12[b] | 55.38±4.51[b] | 34.23±2.40[b] | 28.21±0.31[b] |
| IV-Test1 | 95.63±4.21[c] | 96.58±3.65[c] | 25.68±1.24[c] | 45.33±2.54[c] |
| V -Test2 | 83.23±3.56[b] | 75.22±2.53[b] | 29.47±2.19[b] | 33.54±3.14[b] |
| VI -Test3 | 71.54±2.65[b] | 52.97±1.21[b] | 32.11±2.85[b] | 29.14±2.33[b] |

*Values are expressed as mean ± SD, n=6. [a]P<0.001 values-compared to normal control. [b]P<0.001, [c]P<0.01, [d]P<0.05 values-compared to toxic control.*

**Table 4: Effect of AFME on oxidative stress parameters of liver tissue protein in INH and RIF induced hepatotoxicity**

| Groups | CAT(U/mg) | SOD(U/mg) | GSH(mM/mg) | LPO(mM/mg) |
|---|---|---|---|---|
| I-Normal | 15.75±0.12 | 13.54±0.15 | 6.55±1.02 | 2.54±0.03 |
| II -Toxic | 3.35± 0.31[a] | 5.25±0.21[a] | 1.63±0.42[a] | 5.12±0.12[a] |
| III-Standard | 14.58±0.24[b] | 11.63±0.08[b] | 5.95±0.11[b] | 2.98±0.02[b] |
| IV-Test1 | 6.74±0.03[c] | 7.56±0.51[c] | 3.24±0.09[c] | 4.33±0.14[c] |
| V -Test2 | 9.54±0.11[b] | 9.58±0.23[b] | 4.66±0.03[b] | 3.56±0.22[b] |
| VI -Test3 | 12.56±0.45[b] | 10.75±0.33[b] | 5.69±0.21[b] | 3.18±0.04[b] |

*Values are expressed as mean ± SD, n=6. [a]P<0.001 values-compared to normal control. [b]P<0.001, [c]P<0.01, [d]P<0.05 values-compared to toxic control.*

### 3.7. Effect of AFME on histopathological changes

The effect of AFME on histopathological changes in INH and RIF induced hepatotoxicity in rat liver tissue was shown in fig. 1. Liver of normal control rats showed normal structure having central vein, hepatic cells with normal sinusoidal space, liver sections of rats induced with INH and RIF shows abnormal in structure involves degeneration of cells with cytoplasm dissolution, hepatocellular necrosis, cell proliferation, inflammatory infiltrates with central vein congestion and increase in intracellular spaces with fatty deposits. In treated groups according to given doses, Test1 shows mild, Test2 shows moderate and Test3 shows good recovery and protection from structural damage such as hepatocyte regeneration, mild inflammatory infiltrates with less fatty changes, improved hepatic, central vein and sinusoidal dilation compared to standard group of rats showed adequate structure without any abnormality and degenerative changes of cells.

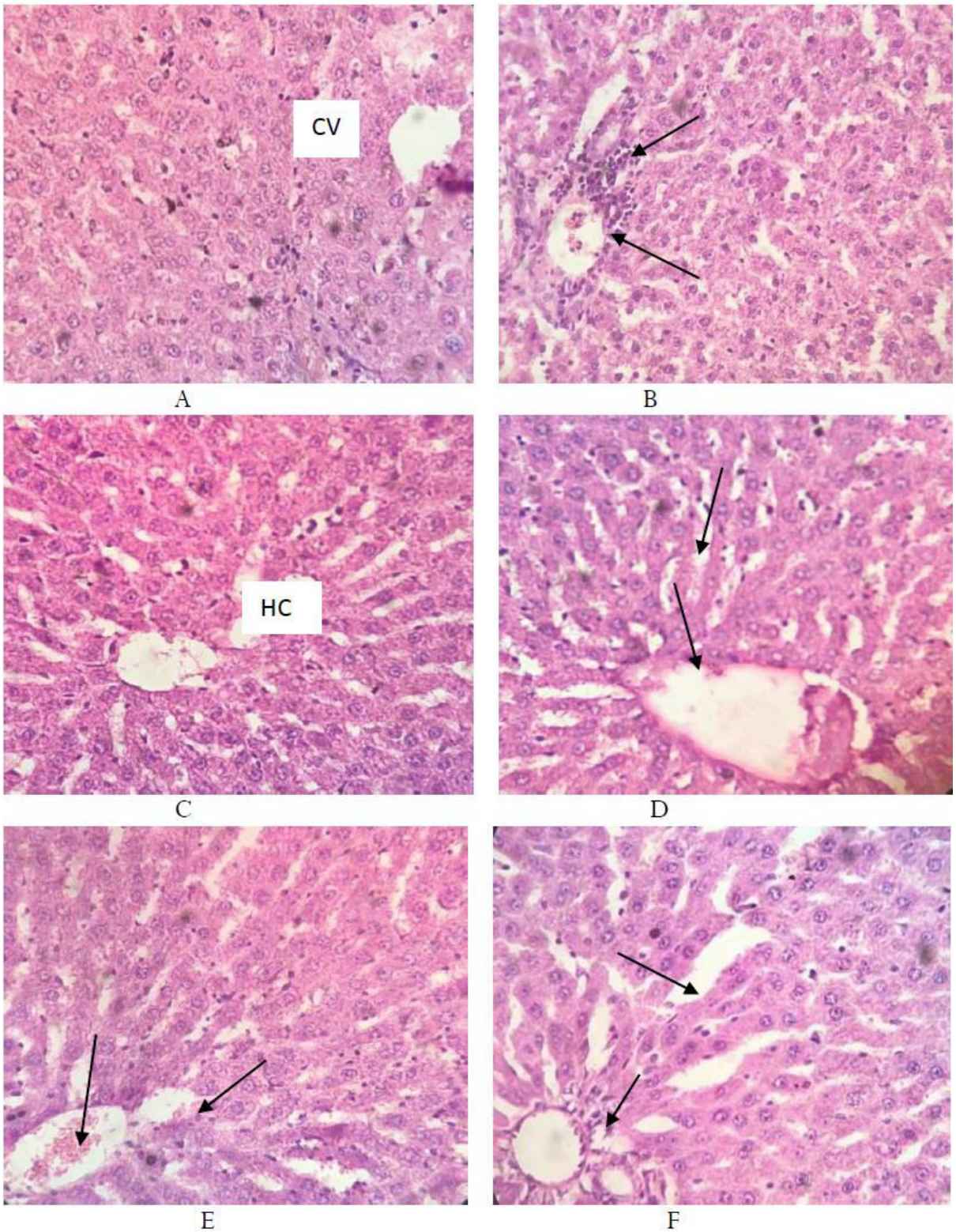


**Fig. 1: Effect of AFME on histopathological changes in INH and RIF induced hepatotoxicity in rat liver tissue**

*Histopathological changes of liver tissue observed under 100X magnification by using Digital Motic Microscope with hematoxylin-eosin stain in normal and treated groups of rats. A. Normal group- showing normal central vein (CV) surrounded with normal hepatic cells (HC). B. Toxic group- showing disorganized structure with hepatic necrosis, cellular infiltration. C. Standard group- showing well organized structure without any structural damage. D,E,F- Test groups 1,2 and 3- showing gradual recovery and mild to moderate protection from structural destruction according to treated doses. Arrow marks indicated the structural damage of liver tissue.*

## 4. DISCUSSION

INH and RIF are well known first line drugs acts on tuberculosis effectively. These drugs not only treat TB but also produce severe hepatic damage. So this study was aimed to screen the hepatoprotective activity of the root of plant *Abelmoschus ficulneus* on drug induced hepatotoxicity caused by INH and RIF combination. Hepatotoxicity produced with INH and RIF administration was proven by the altered levels of serum biochemical parameters, lipid profile parameters, oxidative stress parameters in tissue homogenate along with changes in histopathological observations.

Induced hepatotoxicity with INH and RIF treatment characterized by significantly decreased final body weight of rats associated with abnormal liver enzymes led to impaired metabolism. Significantly increased liver weight and relative liver weight was observed due to accumulation of abnormal cholesterol and triglycerides level leads to significantly altered lipid profile parameters such as elevated levels of cholesterol, triglyceride, LDL-cholesterol and depleted levels of HDL-cholesterol [29]. The increased levels of biochemical parameters AST, ALT, ALP, LDH were noticed significantly, because these are the markers indicate the cellular damage of liver, normally built in cytoplasm and released in to systemic circulation after hepatocellular damage [30]. Increased levels of TB, DB due to accumulation of toxic metabolites [31], decreased TP and ALB levels were recognized due to impaired synthetic functions of liver [32]. Finally, the oxidative stress markers hepatic CAT, GSH, SOD levels were decreased significantly and hepatic LPO levels in tissue homogenate were increased significantly because of an imbalance between oxidant and antioxidant agents [33]. These altered parameters can be allocated to architectural damage of liver. Along with these changes, the changes in histopathological observations like cellular damage, accumulation of fat, structural abnormality confirmed again the hepatotoxicity of INH and RIF.

Treatment of rats with AFME test doses (100, 200 and 400 mg/kg b.wt) along with INH and RIF shows better protection from drug induced hepatotoxicity and it was identified with the gradual improvement of altered parameters such as improved final body weight, significantly decreased liver weight and relative liver weight. The higher levels of serum biochemical parameters AST, ALT, ALP, LDH, TB, DB were diminished and lower levels of TP, ALB were enhanced significantly. The abnormalities in lipid profile were significantly changed in to normal levels. Oxidative stress parameters namely CAT, GSH, SOD levels were improved significantly and increased LPO levels were significantly returned to normal. The recovery from cellular damage, dispersal of fatty deposits, structural normality in histopathological findings revealed that administered AFME gradually protected the rats from hepatotoxicity of INH and RIF and it was well comparable to standard drug silymarin.

The hepatoprotective activity of AFME against drug induced hepatotoxicity was confirmed by dose dependent reversal of biochemical parameters with gradual recovery from histopathalogical abnormalities. This significant hepatoprotection might be possible with the presence of flavonoids, phenolic compounds, alkaloids, glycosides and terpenoids in AFME.

## 5. CONCLUSION

From the results, it was concluded that *Abelmoschus ficulneus* methanolic extract has shown dose dependent hepatoprotective activity against drug induced hepatotoxicity caused by the combined action of isoniazid and rifampicin and the test dose at 400 mg/kg exhibited significantly more effect when compared with doses of 100, 200 mg/kg body weight. Administration of AFME ameliorates the hepatotoxicity by reformation of all altered parameters, improvement of antioxidant defence mechanism along with histopathological changes. And also this study substantiated traditionally claimed health benefit of the plant in treating liver disorders. However further investigation is needed to elucidate the phytoconstituents and mechanism behind the hepatoprotective effect of *A. ficulneus*.

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### *Conflict of interest*

Authors have no conflict of interest.

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