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PHARMACOLOGICAL INVESTIGATIONS OF FICUS LONGIFOLIA SABRE TREE

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ABSTRACT

The goal of this research investigation was to conduct pharmacological research on *Ficus longifolia* for a number of different pharmacological studies. Flavanoids, steroids, terpenes, phenolics, glycosides, alkaloids, amino acids, and reducing sugars were found to be phytoconstituents in portions of the sabre tree. EELFL, EEAPFL, and EERFL had a very large safety margin and were categorised as non-toxic constituents. Significant anti-hepatotoxic action was obtained by EELFL, however it was smaller than that of normal silymarin. EELFL was found to reduce thiopentone sodium-induced sleeping time compared to CCl_4 treatment. Compared to EEAPFL and EERFL, EELFL produced better action and reduced blood glucose. EELFL also markedly reduced blood glucose. Serum cholesterol levels were reduced with EETOL therapy (hypolipidemic / hypocholesterolemic action). It was therapeutic when EELFL raised the HDL level in the treated animals. LDL and VLDL levels were reduced with EELFL (therapeutically beneficial). It is suggested that more study be done to identify and define the pharmacologically active components of the *Ficus longifolia* extracts EELFL, EEAPFL, and EERFL.

KEYWORDS: Ethanolic extract of leaves of *Ficus longifolia* (EELFL), ethanolic extract of aerial parts of *Ficus longifolia* (EEAPFL), and ethanolic extract of roots of *Ficus longifolia* (EERFL).

INTRODUCTION

Liberty *et al*., 1976, genus *Ficus* (diploid; Moraceae) is one of the largest genera (with more than 850 distributed throughout the tropical and sub-tropical regions of the world) of angiosperms species. *Ficus* species contains woody trees, shrubs, vines, epiphytes and hemiepiphytes. (Wagner and Zotz, 2020)

Gotsch *et al*., 2015, *Ficus longifolia* (Sabre Tree, Alii Tree, Amstel King, Fig Tree; evergreen; ornamental) indoor plant tree with narrow leathery leaf, medium tree, on arching branches, mahogany trunks speckled with white bars. *Ficus* consists of vast amounts of sap in bark, branching, leaves (linear-lanceolate laminas, narrow laminas, long stipules; 10-25 x 1.5-3.5 cm), flowers are tiny (syconium fig), 3-6 mm across, whitish to pinkish. (Figure 1)

Figure 1: Field Photograph and Leaves of *Ficus longifolia*.

Ficus longifolia contains phenolics, sterols, coumarins and / or furano-coumarin, chromone, triterpenes, glycosides, isoflavones and lignans; mangiferin; catechin and epicatechin; rutin; acids like 7-hydroxycoumeric, vanillic, luteolin, quercetin, apigenin, kaempferol, chrysin, ursolic, oleanolic.

Ficus longifolia is traditionally used as a mild laxative, anti-rheumatic, galactagogue, digestive and as anthelmintic against intestinal parasites.

Pharmacological activities of various *Ficus* species include antioxidant activity (Akhtar *et al*., 2019), anti-microbial activity (Raja *et al*., 2021), anti-inflammatory and analgesic activities (Li *et al*., 2021; Kumar *et al*., 2021), Anti-malarial (Ifijen *et al*., 2020); anti-ulcer actions (Fokunang *et al*., 2019), Anti-fungal (Chen *et al*., 2020) Anti-HIV (Indriati *et al*., 2020), wound healing (Manjuprassana *et al*., 2020) etc.

Experimental / Methods

Collection and Authentication of *ficus longifolia*

In September 2021, fresh *Ficus longifolia* (sabre tree) plant components (leaves, aerial parts, and roots) were harvested from the Glocal University campus's Medicinal Plant Garden. Pharmacognostically, the leaves of the Sabre tree were examined to ensure authenticity. Herbarium specimens (GU/GSOP/Pharm/Herb/2021/107) was also deposited.

Phytochemical screening of *ficus longifolia*

Using standard procedures outlined by Harborne (1973), Trease and Evans (1985), Sofowora (1993), Khandelwal (2008), Kokate (2005), and others, the petroleum ether, chloro -form, methanol, and aqueous extracts of the plant aerial parts, leaves, and roots (dried under shade, coarsely pulverised separately) of the sabre tree were subjected to preliminary phytochemical screening for PPMs and SPMs. Qualitative analysis of sabre tree plant parts showed the presence of PPMs and SPMs like glycosides; alkaloids; amino acids; flavanoids; steroids; terpenes, phenolics; and reducing sugars.

Preparation of ethanolic extract

Shade from sabre trees aerial parts, leaves, and roots (1000 gm each) were dried and coarsely ground. They were then Soxhlet extracted for 16 hours using 80% crude et hanol, they were then filtered, distilled, vacuum evaporated, and lyophilized.Extracts with pra ctical yields for EEAPFL, EELFL, and EERFL were 1.12%, 1.86%, and 1.54%, respectively. Pharmacological studies employed varying doses of EEAPFL, EELFL, and EERFL extracts (1% carboxymethylcellulose).

Anti-hepatotoxic activity

IAEC of Glocal School of Pharmacy, Glocal University (CPCSEA guidelines) approved Form-B GU/GSOP/IAEC/2023/11 (17-02-2023). Animals were given water *ad libitum* and on eighth day liver biochemical parameters (LFTs) were estimated. The blood samples for the LFT estimations were collected from retro-orbital sinus. Animals of Group II were served as toxic control group (CCl_4 Group) and animals were given Carbon-tetrachloride (1.5 ml/kg p.o.) single dose. Group III to IX animals were given CCl_4 (1.5 ml/kg) in single dose to induce hepatotoxicity followed by treatment with EELFL (Group III; 200 mg/kg), EELFL (Group IV; 400 mg/kg), EEAPFL (Group V: 200 mg/kg), EEAPFL (Group VI: 400 mg/kg), EERFL (Group VII: 200 mg/kg), EERFL (Group VIII: 400 mg/kg) for 03 weeks and standard drug Silymarin (140 mg/kg; Group IX). SGPT / ALT, SGOT / AST, Abumin, Total proteins

(T-Prot), alkaline phosphatase (AKLP) and billirubin were estimated (Gornal *et al*., 1949; Lowry *et al*., 1949; Godfried *et al*., 1935).

Preparation of Formalin - Propionic acid – Alcohol (FPA) Solution

Formalin – propionic acid – alcohol (FPA) is used for chemical fixation of biological tissues. It comprises of Formaldehyde (fixing agent), Propionic acid (non-coagulated fixing agent) and Ethanol (coagulant fixing agents). Its composition is as follows (Table 1):

Table 1: Composition of Formalin- Propionic acid-Alcohol (FPA) Solution.

S. No.	Component	Quantity (ml)
1	Formalin (Formaldehyde 37 %)	10
2	Propionic acid	05
3	Alcohol (95%)	50
4	Distilled Water	35

FPA fixative solution penetrates tissue quickly and stablises them. The tissue is stabilized by the cross-linking property of formaldehyde. Fixed tissues were subjected to microtomy technique stepwise (i) Washing, (ii) Dehydration, (iii) Infiltration, (iv) Embedding, (v) Sectioning. Tissues taken from FAA / FPA solution and TS livers sections were observed for histo-pathological changes.

Effects of Extracts on Thiopentone sodium induced Sleeping Time and Liver Weight Analysis

Kulkarni, 1999 reported that barbiturates are extensively metabolized in the liver and damaged liver suffers with delay in barbiturates clearance (thiopentone sodium 40 mg/kg i.p; cause longer duration of hypnotic effect; Gujrati *et al*., 2007). Group I animals were given water *ad libitum* / vehicle control. Group II animals were Toxic control animals were given CCl_4 (1.5 ml/kg). Animals of Group II to VI were served as models for CCl_4 (1.5 ml/kg) induced hepatotoxicity. Animals of Group II to V were given CCl_4 (1.5 ml/kg) to induce hepatotoxicity followed by treatment with EELFL / EEAPFL / EERFL (400 mg/kg) with Thiopentone sodium (40 mg/kg i.p) (Group III, IV and V) and Silymarin (standard drug, 140 mg/kg) with Thiopentone sodium (40 mg/kg i.p) (Group VI).

Anti-diabetic Activity of EELFL, EEAPFL and EERFL against Streptozotocin

IAEC approved Form-B proposal GU/GSOP/IAEC/2023/11 on 17-02-2023.

Effect of "EETOL", EETOP and "EETOR" on Glucose level

Animals were treated with EELFL, EEAPFL and EERFL and the normal group received water. Animals were anaesthetised with Ketamine+Xylazine ((80 mg/kg+10 mg/kg i.p.) and blood was withdrawn from retro orbital plexus and blood glucose levels were estimated with Glucometer or by GOD/POD method at 1, 2, 3 h.

Effect of EETOL, EETOP & EETOR on "Oral Glucose Tolerance Test"

Oral glucose tolerance test (OGTT) was estimation was carried out in overnight fasted normal rats. Animals were administered EELFL (400 mg/kg), EEAPFL (400 mg/kg) and EERFL (400 mg/kg). Glucose (2 g/kg b.w.) was given orally (60 min after dosing) and glucose levels were estimated.

Comparison of anti-diabetic effects of EELFL, EEAPFL, EERFL

STZ in 0.1M citrate buffer (pH 4.5) was given to animals and glucose levels were analyzed after 07 days. Animals were treated with EELFL, EEAPFL and EERFL (Group III-VIII) and Glipizide (Standard, Group IX) as prescribed and after 21 days blood glucose levels were analysed.

Effect of EELFL, EEAPFL and EERFL on Lipid Profile in diabetic rats

Total cholesterol: Cholesterol estimated with Agappe Liqui CHECK kit.

Estimation of triglycerides (mg /dl)

$$\text{Triglycerides in (mg /dl)} = \frac{\text{Absorbance of sample}}{\text{Absorbance of standard}} \times 200$$

Where, 200 is the standard concentration

High density lipoprotein cholesterol: HDL–C levels estimated (Crest Bio systems).

High Density Lipoprotein (LDL): Friedwald *et al.*, 1972 method was used:

LDL cholesterol = TC – (VLDL + HDL cholesterol)

VLDL: Friedewald *et al.*, 1972 illustrated VLDL visualized by formula:

VLDL cholesterol = TG/5

RESULTS

To ensure authenticity and purity, fresh plant sections from the Sabre tree were examined morphologically, microscopically, and chemically (to find PPMs and SPMs). Sabre tree plant sections revealed the presence of flavanoids, steroids, terpenes, phenolics, glycosides, alkaloids, amino acids, and reducing sugars. A herbarium specimen (GU/GSOP/Pharm/Herb/2021/107) was deposited.

The administration of CCl_4 (1.5 ml/kg) single dose induced severe liver damage (manifested in 24 hrs of ingestion of toxic dose) which include glutathione depletion, necrosis of hepatocytes, elevated levels of plasma amino-transferases, and increased concentration of billirubin. Biopsy of the showed centilobular necrosis (which include necrosis, degeneration, and infiltration) and rise in the level of LFTs were correlated with the hepatic lesions produced. (Figure 3)

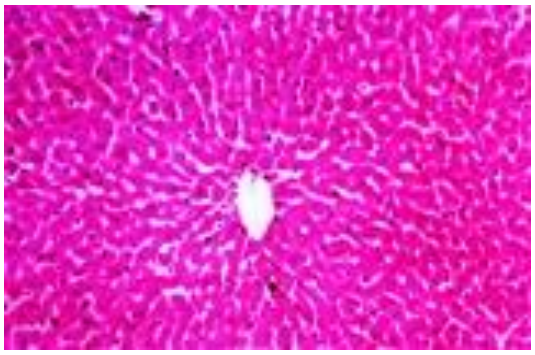

Figure 2: TS of Normal Liver.

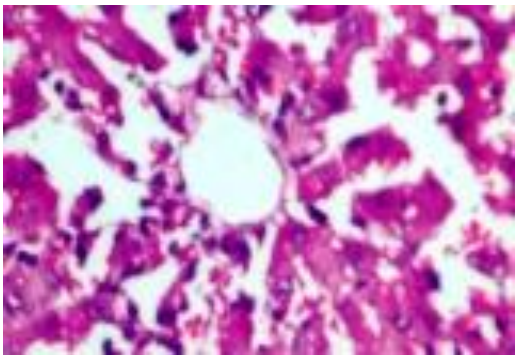

Figure 3: CCl_4 induced toxic liver.

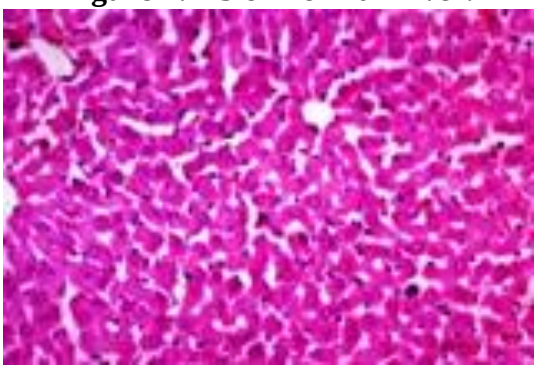

Figure 4: EELFL (400 mg) liver.

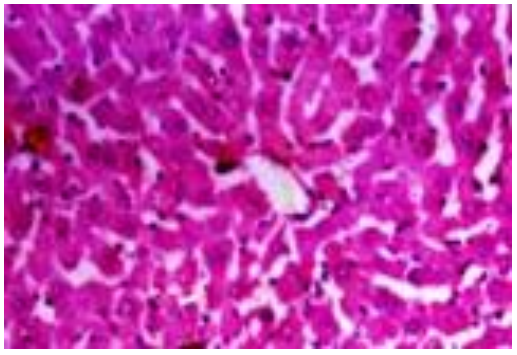

Figure 5: Standard drug liver.

Administration of EELFL extract (400 mg/kg) for 02 week (Group IV) produced significant alleviation of the serum enzyme activities. EELFL produced significant reversal effects which were clearly indicated by reddish coloration, normal anatomical architecture of the hepatocytes. EELFL showed a remarkable anti-hepatotoxic activity (Table 2; Figure 4) but comparatively lesser then standard reference drug i.e. Silymarin (Group IX; Figure 5 & 6)

and data represented in Table 2 was analyzed by ANOVA.

Table 2: Effects of EELFL, EEAPFL and EERFL on LFTs in wistar rats.

Group	SGPT (Units/ml)	SGOT (Units/ml)	Serum albumin	Total Protein	Serum alkaline phospha-tase	T. Bil. (mg/dl)
Group I (Normal) Fig. 2	34.4 ± 2.42	38.8 ± 2.24	3.56 ± 0.36	4.78 ± 0.32	18.14 ± 1.44	0.24 ± 0.14
Group II (CCl_4 Control) Fig. 3	186.8 ± 4.42	196.4 ± 2.84	3.92 ± 0.44	4.94± 0.46	34.4 ± 2.3	0.86 ± 0.16
Group III *(EELFL; 200 mg/kg)*	136.4 ± 2.8	166.8 ± 2.34	3.64 ± 0.38	4.90 ± 0.28	27.6 ± 1.46	0.66 ± 0.18
Group IV (EELFL; 400 mg/kg) Fig. 4	114.4 ± 2.4*	124.6 ± 2.16*	3.62 ± 0.32*	4.88 ± 0.24*	25.8 ± 1.32*	0.64 ± 0.14*
Group V *(EEAPFL; 200 mg/kg)*	156.2 ± 2.34	168.4 ± 1.52	3.72 ± 0.28	4.90 ± 0.18	27.6 ± 0.42	0.74 ± 0.62
Group VI (EEAPFL; 400 mg/kg)	126.6 ± 2.24	138.8 ± 1.56	3.68 ± 0.28	4.82 ± 0.16	26.4 ± 0.44	0.68 ± 0.26
Group VII (EERFL; 200 mg/kg)	168.8 ± 3.52	174.2 ± 2.46	3.86 ± 0.24	4.90 ± 0.36	31.4 ± 3.18	0.82 ± 0.26
Group VIII (EERFL; 400 mg/kg)	148.8 ± 3.52	152.8 ± 2.12	3.86 ± 0.42	4.84 ± 0.36	29.4 ± 3.42	0.76 ± 0.26
Group IX (Silymarin) Fig. 5	78.6 ± 2.36*	87.6 ± 1.42*	3.56 ± 0.34*	4.64 ± 0.34*	22.8 ± 0.64*	0.36 ± 0.14*

Note: *P<0.05 (Significance difference)

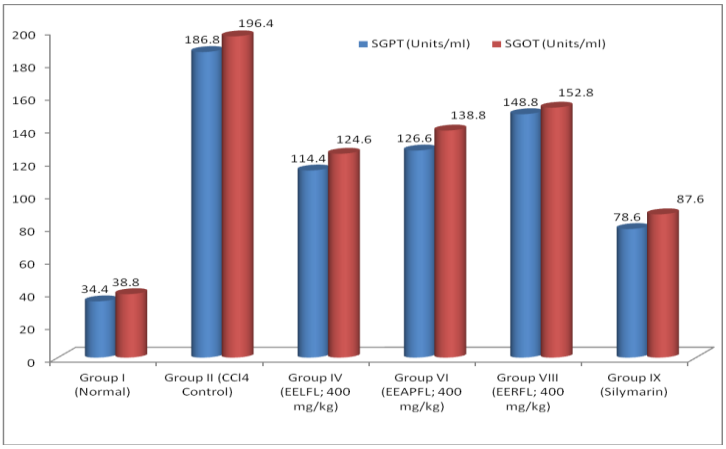

Figure 6: Effects of EELFL, EEAPFL, EERFL & Silymarin on SGPT & SGOT.

Table 3: Effect of Extracts on thiopentone induced sleeping time & liver weight.

Group	Thiopentone sod. induced sleeping time		Liver wt (g/100g bw)
	Onset (s)	Duration (min)	
Group I (Normal control)	206.54 ± 4.82	76.54 ± 4.72	3.92 ± 0.50
Group II (Toxic Control)	52.34 ± 4.22a	244.32 ± 4.62a	6.46 ± 0.30a
Group III (EELFL 400 mg/kg)	154.42 ± 4.46***	132.82 ± 5.84***	5.24 ± 0.12*
Group IV (EEAPFL 400mg/kg)	132.62± 4.44	156.28 ± 5.26	5.68 ± 0.14
Group V (EERFL 400 mg/kg)	64.24 ± 4.18	216.14 ± 4.68	6.28 ± 0.18
Group VI (Silymarin)	182.28 ± 6.46***	120.26 ± 5.24***	4.84 ± 0.16*

Values are mean ± SEM of 6 animals in each group

aP < 0.001 relative to control group;

***P < 0.001 relative to Toxicant group

*P < 0.05 relative to Toxicant group

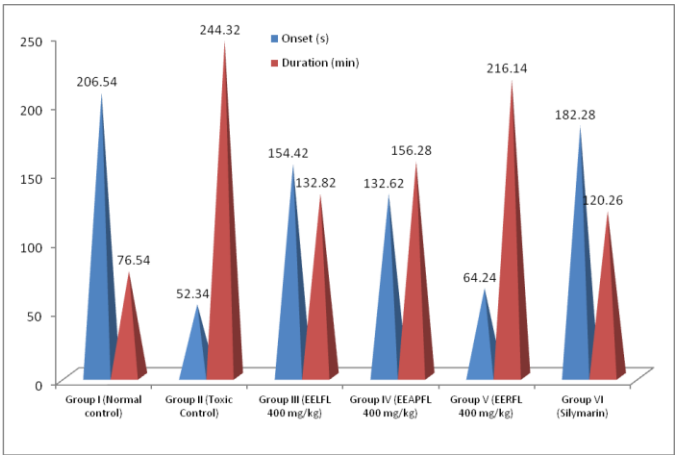

Figure 7: Effect of Thiopentone sodium on onset and duration of sleeping time.

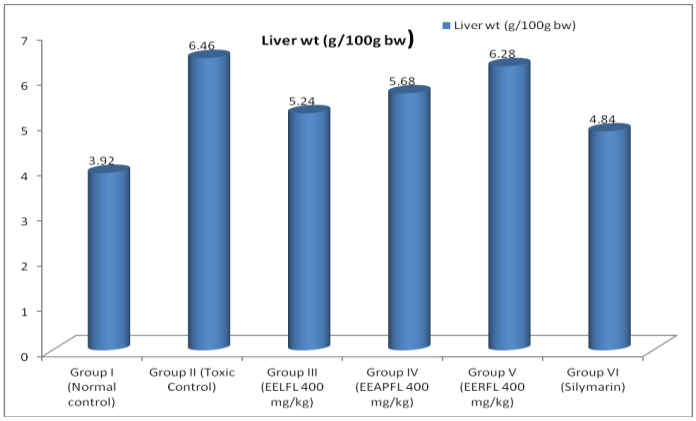

Figure 8 : Effect of Thiopentone sodium on Liver Weight (g/100g bw).

Sleeping time / pattern were normal in Group-I animals. Administration of CCl_4 (1.5 ml/kg) in single dose induced severe liver damage. Subsequently, CCl_4 induced hepatotoxicity exhaust glutathione stores and leads to necrosis of hepatocytes and liver weight were also increased in toxic control group animals. Reduction in thiopentone sodium induced sleeping-time was recorded with EELFL when compared to CCl_4 treated (Table 3; Figure 7 and 8).

Sabre tree EELFL has induced glucose lowering effect after 2 hr. Besides, sugar levels were restored after 3 hr in all treatment groups (Table 4). EELFL had alleviated blood glucose (maximum reduction after 21 days) and produced better action than EEAPFL and EERFL ($P < 0.001$; Table 4-6; Figure 9-11).

Table 4: Normoglycaemic effects of EELFL, EEAPFL, EERFL.

Groups	Blood glucose (mg/dl)			
	0 hr	1 hr	2 hr	3 hr
Control (DW)	76.2 ±.2	75.4 ±.2	73.6 ±.2	72.6 ±.8
EELFL (400 mg/kg)	78.2±.4	74.2±.2	72.6±.8	74.6±.4
EEAPFL (400 mg/kg)	74.6±.4	76.2±.2	80.2±.8	78.8±.2
EERFL (400 mg/kg)	75.4±.4	78.2±.4	83.4±.6	82.6±.6

Note: Mean ± SEM (triplicate measurements)

Table 5: EELFL, EEAPFL and EERFL effect on glucose tolerance.

Groups	Blood glucose (mg/dl)				
	0 min	30 min	60 min	90 min	120 min
Control	80.6±.42	138.2±.42	164.8±.22	138.6±.22	128.3±.32
EELFL (400 mg/kg)	77.4±.20	127.26±.22	112.4 ±.18	96.3±.28	84.2±.28
EEAPFL (400mg/kg)	82.6±.64	128.6±.24	122.6± .32	108±.1.8	102.8±.24
EERFL (400 mg/kg)	82.4±.64	130.2±.32	124.2± .30	112±.2.6	106.8±.32

Note: Mean ± SEM.

Table 6: EELFL, EEAPFL and EERFL effects on blood glucose against STZ.

Group	Blood Glucose (mg/dl)			
	Day 1	Day 7	Day 14	Day 21
Control	92.2 ± 4.34	95.2 ± 3.84	94.4 ± 3.92	94.8 ± 4.22
Toxic Control	212.4 ± 6.34	206.4 ± 5.72	202.6 ± 6.14	198.4 ± 4.32
EELFL	206.2 ± 4.36	170.4 ± 3.84	134.6 ± 4.14	108.6 ± 2.86
EEAPFL	204.6 ± 8.24	194.2 ± 4.56	148.6 ± 4.36	132.2 ± 4.32
EERFL	204.2 ± 6.26	182.4 ± 3.52	162.6 ± 3.52	154.8 ± 4.64
Glipizide	206.4 ± 5.22	162.2 ± 3.84	124.6 ± 3.24	98.4 ± 2.72

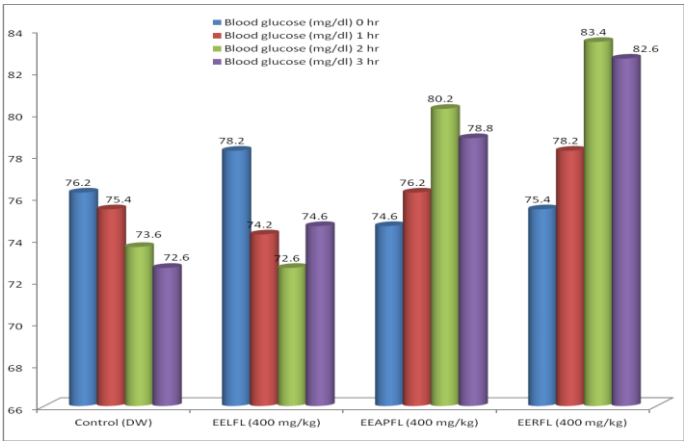

Fig. 9: Effect of EELFL, EEAPFL & EERFL on glucose in normo-glycaemic rats.

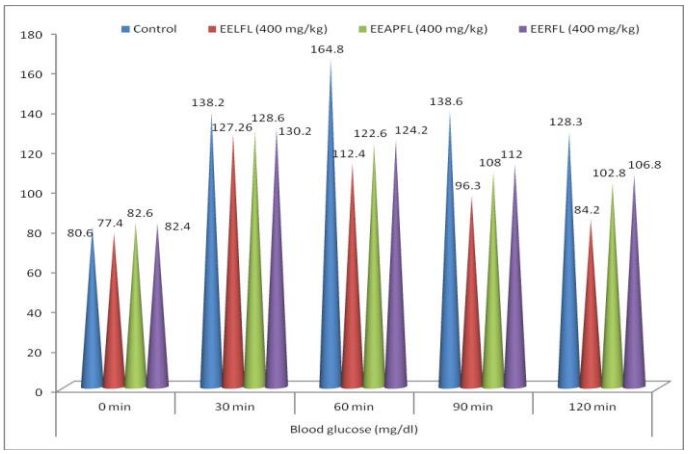

Figure 10: EELFL, EEAPFL and EERFL effects on glucose-tolerance.

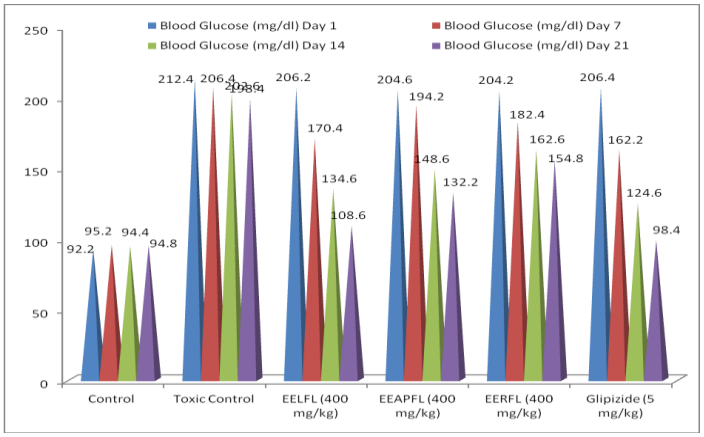

Figure 11: Anti-diabetic effects of extracts and Glipizide (Std.) against STZ.

Further, EELFL (400 mg/kg) lowered blood glucose significantly from 206.2 (1st day), 170.4 (7th day), 134.6 (14th day) to 108.6 (21st day) (Table 4.23; Figure 4.20). On the other hand blood glucose in animals treated with EEAPFL (400 mg/kg) was 204.2 (Day-1), 182.4 (day-7), 162.6 (day -14) and 154.8 on day-21. Glipizide (standard drug) significantly lowered glucose level from 206.4 (day 1)-162.2 (day -7)-124.6 (day-14) to 98.4 mg/dl after 21 days.

Results recorded in Table 7 (Figure 12) indicated the effect of EELFL, EEAPFL and EERFL on cholesterol and triglycerides level in STZ induced diabetic rats. STZ toxicity induced increase in serum cholesterol (after 7 days 266.4; after 21 days 221.6), triglycerides [200.2 (7 days); 174.8 (21 days)] ($P < 0.0001$). EETOL treatment alleviated the level serum cholesterol [142.4 (7days); 120.0 (21 days)], triglycerides [132.4 (7 days); 84.4 (21 days)]. Treatment with EEAPFL alleviated cholesterol [154.6 (7days); 134.6 (21 days)], triglycerides [148.6 (7 days); 92.4 (21 days)].

Standard reference drug glipizide (5mg/kg) decreased cholesterol the [112.6 (7 days); 98.4 (21 days)], triglycerides [98.6 (7 days); 88.6 (21 days)]. EELFL has prominent hypolipidemic / hypocholesterolemic activity but lesser than Standard drug ($P < 0.0001$).

Table 7: Effect of EELFL and EEAPFL on serum cholesterol and triglycerides.

Groups	After 7 days		After 21 days	
	Cholesterol	Triglyceride	Cholesterol	Triglyceride
Control	114.2± 1.4	102.2±.2.8	108.6±1.26	98.8±2.24
Diabetic control	266.4±2.4	200.2±1.2	221.6±2.84	174.8±2.4
EELFL (400mg/kg)	142.4±2.4	132.4±1.54	120.0±1.82	84.4±2.26
EEAPFL (400mg/kg)	154.6±2.8	148.6±2.24	134.6±1.26	92.4 ± 2.14
EERFL (400mg/kg)	184.4±2.8	168.2±2.24	154.8±1.64	142.6 ± 2.14
Glipizide (5mg/kg)	112.6±1.52	98.6±1.28	98.4±1.24	88.6 ± 1.12

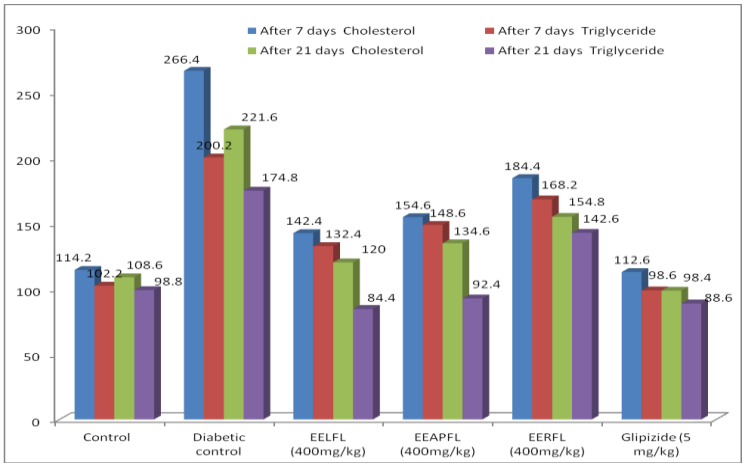

Figure 12: Effect of Extracts on cholesterol and triglycerides in diabetic animals.

Table 8: Effect of EELFL, EEAPFL, EERFL and Glipizide on lipoproteins.

Groups	After 7th day			After 21st day		
	HDL	LDL	VLDL	HDL	LDL	VLDL
Control	60.4±0.4	32.4±0.46	20.6±0.14	40.4±0.22	36.0±.64	14.4±.02
Toxic	21.2±0.2	192±2.42	41.2±0.12	27.2±1.44	134.4±1.2	40.2±.16
EELFL	34.4±.16	80.6±.84	26.4±.16	42.6±1.6	58.2±1.2	15.6± 0.4

EEAPFL	32.4±.22	91.2±.32	27.6±.12	43.2±1.2	62.4±1.84	18.8±0.12
EERFL	28.8±.24	96.4±.36	27.2±.18	36.4±1.4	66.2±1.76	18.2±0.14
Glipizide	38.6±.26	72.8±.74	24.2±.32	46.8±1.6	48.6±1.24	14.2± 0.6

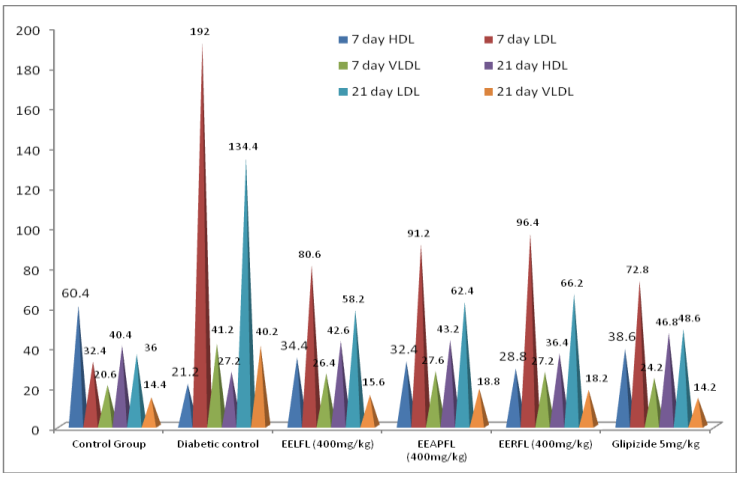

Figure 13: Effect of extracts on lipoproteins in STZ induced diabetic rats.

In normal animals HDL (good cholesterol) level were found to be 60.4 (7 days) and 40.5 (after 21 days). In diabetic control / toxic control group HDL level were 21.2 (7 days) and 27.2 (21 days).

HDL was significantly lowered in diabetic rats when compared to normal control ($P < 0.0001$). EELFL restored HDL level 34.4 (7 days) and 42.6 (21 days) and EEAPFL treatment the produced HDL level 32.4 (7 days) and 43.2 (21 days). In glipizide (5mg/kg) treated group serum HDL was 38.6 (7 days) and 46.8 (21 days). Beneficial effect of EELFL and EEAPFL on HDL level in treated animals was therapeutic (Table 8; Figure 13).

In healthy animals (normal group) LDL (bad cholesterol) level were found to be 32.4 (7 days) and 36.0 (after 21 days). In toxic control group LDL level were 192 (7days) and 134.4 (21 days). LDL was significantly increased in STZ toxic group animals ($P < 0.0001$). EELFL (400 mg/kg) alleviated LDL level 80.06 (7 days) and 58.2 (21 days) and EEAPFL (400 mg/kg) treatment the produced LDL level 91.2 (7 days) and 62.4 (21 days). In glipizide treated group serum LDL was 72.8 (7days) and 48.6 (21 days). Beneficial effect (LDL alleviating effect) of EELFL (400 mg/kg), EEAPFL (400 mg/kg) and Standard on LDL level in treated groups was good (Table 8; Figure 13).

In normal group VLDL level were found to be 20.6 (7 days) and 14.4 (21 days). In diabetic toxic control group VLDL were 41.2 (7days) and 40.2 (21 days). EELFL (400 mg/kg) alleviated VLDL level 26.4 (7 days) and 15.6 (21 days) and EEAPFL (400 mg/kg) treatment

the produced VLDL level 27.6 (7 days) and 18.8 (21 days). In glipizide treated group serum VLDL level was 24.2 (7days) and 14.2 (21 days). VLDL alleviating effect of EELFL (400 mg/kg) and EEAPFL (400 mg/kg) on LDL level group III-IV were therapeutically positive (Table 8; Figure 13)**.**

CONCLUSIONS

It was concluded that EELFL, EEAPFL, and EERFL had a very large safety margin and were categorised as non-toxic constituents (no mortality even at doses of 2000 mg/kg b. wt.) and safe for pharmacological use.

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REFERENCES

1. Akhtar, P., Yaakob, Z., Ahmed, Y. *Turk. J. Pharm. Sci*, 2019; 2: 1-14.
2. Chen, Y., Zhang, M., Zhang, T. *Antioxidants*, 2020; 11: 942.
3. Fokunang, E. T., Pougoue, J. K., Fokunang, C.N. *Intl. J. Biol. Chem. Sci*, 2019; 13(1): 475-492.
4. Friedewald, W. T., Levy, R. I., Fredrickson, D. S. *Clin Chem*, 1972; 18(6): 499-502.
5. Godfried, *Biochem. J*., 1935; 29: 1337.
6. Gornall, A.C., Bardawill, C.J., and David, M.M. *Journal of Biological Chemistry*, 1949; 177: 751-767.
7. Gotsch, S., Darbylx, A., Dawson, T. *Ecol. Monogr, 2015;* 85(3): 393-412.
8. Gujrati, V., Rao, V., Nandakumar, K., *Ind. J. Pharmacol*. 2007; 39: 43-47.
9. Harborne, J. B. *A Guide to Modern Techniques of Plant Analysis*. Springer Publications, 1973; 1-288.
10. Ifijen, I.H., Odiachi, I.J., Aghedo, O.N., *Chem. Afr*, 2020; 3(4): 1-7.
11. Indriati, D.W., Tumewu, L., Khairunisa, S.Q. *Res. J. Pharm. Techn, 2020;* 13(1): 1-4.
12. Khandewal, K.R. *Prac. Pharmacognosy*. Nirali, edition, 2008; 19: 1-67.
13. Kokate, C.K. *Pract. Pharmacog*. Vallabh: Delhi, 2005; 5: 107- 111.
14. Kulkarni, S.K. *Hand book of experimental pharmacology*. Vallabh Prakashan: New Delhi, 1999; 3: 1-78.
15. Kumar, S., Gaidhani, S., Sudhakar, D. *World J. Pharm. Res*, 2021; 10(3): 1644-1652.

16. Li. Z., Liu, M., Shao, J., Cui, Q. *Biomed. Pharmacother,* 2021; 137: 1-10.
17. Liberty, H., and Bailey, H. Hortus third: A concise dictionary ofplants cultivated in the USA and Canada. New York, 1976.
18. Lowry, O., Bessel, Crawford, E. *J. Biol. Chem, 1949;* 180: 399.
19. Manjuprasanna, V.N., Rudresha, G.V., Rajaiah, R., Vishwanath, B.S. *Intl. J. Biol. Macromol, 2020;* 165: 691-700.
20. Raja, R., Kumar, N., Duraisamy, M. *Kongunadu Res,* 2021; *J.* 8(1): 22-29.
21. Sofowora, A. J. *Altern. Complement. Med.* 1993; 2(3): 365-372.
22. Trease, G.E., Evans, W.C. Pharmacognosy. Tinal, London, 1985; 149.
23. Wagner, K., Zotz, G. *J. Ecol,* 2020; 108(2): 761-773.