ISSN: 2321-3132

Research Article

International Journal of Chemistry and Pharmaceutical Sciences

www.pharmaresearchlibrary.com/ijcps

Evaluation of antidiabetic activity of methanolic leaves extract of *Lagerstroemia microcarpa* in streptozotocin induced diabetic rats

Revitha. L, C.D. Shaji Selvin*

Sree krishna College of Pharmacy and Research Centre, Parassala, kerala, India

Received: 24 July 2014, Accepted: 27 August 2014, Published Online: 27 October 2014

Abstract

In the present study, the effect of alcoholic leaves extract of *Lagerstroemia microcarpa*on blood glucose,plasma insulin, and carbohydrate metabolic enzymes were studied in experimental diabetes. Diabetesmellitus was induced by a single intraperitoneal injection of STZ (60 mg/kg bw). Five days after STZinduction, diabetic rats received *L. microcarpa* orally at the doses of 200 and 400 mg/kg daily for 3 weeks.Graded doses of leaves extract showed a significant reduction in blood glucose levels and improvement inplasma insulin levels. The effect was more pronounced in 200 and 400 mg/kg. *L. microcarpa*showed significant increase in hexokinase, Glucose-6-phosphate dehydrogenase and glycogen contentin liver of diabetic rats while there was significant reduction in the levels of glucose-6-phosphataseand fructose-1,6-bisphosphatase. The present study clearly indicated significant antidiabetic effect withthe leaves extract of *L. microcarpa*and lends support for its traditional usage.

Key words: antidiabetic, *In-vitro* antioxidant, *Lagerstroemia microcarpa*, methanolic extract

Contents

***Corresponding author**
C.D. Shaji Selvin
Sreekrishna College of Pharmacy and Research Centre, Parassala, kerala, India
Manuscript ID: IJCPS2218

PAPER-QR CODE

1. Introduction

Diabetes is a major health problem affecting major populations worldwide. It is a chronic disorder in metabolism of carbohydrates, proteins, and fat due to absolute or relative deficiency of insulin secretion with/without varying degree of insulin resistance. There are more than 30 million people with diabetes mellitus in India and the incidence is increasing. Also, there are many patients in the community with undiagnosed diabetes. Decreased physical activity, increasing obesity, stress and changes in food consumption have been implicated in this increasing prevalence in the past two decades. Diabetes is being projected as the World's main disabler and killer in the next 25 years[1]. Epidemiological studies and clinical trials strongly support the notion that hyperglycemia is the principal cause of complications. Effective blood glucose control is the key for preventing or reversing diabetic complications and improving quality of life in patients with diabetes. Thus sustained reduction in hyperglycemia will decrease the risk of developing microvascular complications and most likely reduce the risk of macrovascular complications [2].

Therefore, the present work was undertaken to explore the antidiabetic activities of leaves of methanolic extract of *Lagerstroemia microcarpa* in streptozotocin (STZ) induced diabetic Wistar rat. *Lagerstroemia microcarpa* Family: Lythraceae Common name: Ben Teak Common in deciduous and semi-evergreen forests and found occasionally along margins and opening of evergreen forests, up to 1000 m, endemic to Western Ghats, throughout all district of Kerala, Kanyakumari etc. The *Lagerstroemia microcarpa* species is used in the treatment of Asthma, Chronic Bronchitis, cold, cough and local application for aphathae of the mouth. Seed have been documented for its multiple pharmacological activities including narcotic principal. The leaves were also evaluated for potent, anti-inflammatory and antipyretic activities. However, no data are available in the literature on the antidiabetic and antioxidant activity of leaves of *Lagerstroemia microcarpa*. Therefore we undertook the current investigation to examine the total phenolic content, antioxidant activities of methanolic extract from leaves of *Lagerstroemia microcarpa* through various *in vitro* models.

2. Materials and Methods

Plant Material

The leaves of *Lagerstroemia microcarpa* was collected from Kottoor forest Trivandrum district, Kerala, India. Taxonomic identification was made from Botanical Survey of Medicinal Plants Unit Siddha, Government of India, Palayamkottai. The leaves of *Lagerstroemia microcarpa* was dried under shade, segregated, pulverized by a mechanical grinder and passed through a 40 mesh sieve. The powdered plant materials were stored in an airtight container.

Preparation of *Lagerstroemia microcarpa* extract

The leaves of *Lagerstroemia microcarpa* was dried in shade and powdered. The powdered plant materials were successfully extracted with methanol extraction (80ºC) by hot continuous percolation method in Soxhlet apparatus[3]for 24 hrs. The solvent from the extracts was recovered under reduced pressure using rotary evaporator and subjected to freeze drying in a lyophilizer until dry powder was obtained.

Animals and Diet

16-19 weeks-old adult male Wistar rats, weighing approximately 150 to 200g, were acclimatized for 7 days at temperature (25±2°C) and relative humidity (55±1%) in a 12-hour light/dark cycle in a room under hygienic condition. They were given access to water and fed with standard pellet diet *ad libitum.* The experiments were carried out as per the guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), New Delhi, India, and approved by the Institutional Animal Ethics Committee (IAEC), (Approved number: SKCPRC/2013-2014/IAEC/07).

Drugs and Chemicals

All the biochemicals used in this experiment were obtained from Sigma Chemical Company (St. Louis MO, USA). All other chemicals utilized were obtained either from Hi Media (Mumbai) or SD-Fine Chemicals (Mumbai). All chemicals used wereof analytical grade.

Preliminary phytochemical screening of methanolic extracts of leaves of *Lagerstroemia microcarpa*

The methanolic extract of leaves of *Lagerstroemia microcarpa* was subjected to the following chemical tests separately for the identification of various active constituents[4].

Evaluation of in-vitro antioxidant activity of methanolic extract of leaves of *Lagerstroemia microcarpa*

DPPH photometric assay[5]

A methanolic solution of 0.5ml of DPPH (0.4mM) was added to 1ml of the different concentrations of plant extract and allowed to react at room temperature for 30 minutes. Methanol served as the blank and DPPH in methanol without the extracts served as the positive control. After 30 min, the absorbance was measured at 518nm and converted into percentage radical scavenging activity as follows.

$$Scavenging activity(\%) = \frac{A_{518}\ \text{Control} - A_{518}\ \text{Sample}}{A_{518}\ \text{Control}} \times 100$$

Where A_{518} control is the absorbance of DPPH radical with methanol; A_{518} sample is the absorbance of DPPH radical with sample extract/ standard.

Hydroxyl radical scavenging activity [6]

The assay is based on quantification of degradation product of 2-deoxy ribose by condensation with TBA. Hydroxyl radical was generated by the Fe^{3+}-Ascorbate-EDTA-H_2O_2system (Fenton reaction). The reaction mixture contained 0.1 ml deoxyribose (2.8mM),0.1 ml EDTA (0.1 mM), 0.1 ml H_2O_2 (1mM), 0.1 ml Ascorbate (0.1mM), 0.1 ml KH_2PO_4-KOH buffer, P^H 7.4 (20mM) and various concentrations of plant extracts in a final volume of 1 ml. The reaction mixture was incubated for 1 hour at 37°C. Deoxyribose degradation was measured as TBARS and the percentage inhibition was calculated.

Ferric Reducing Ability Power (FRAP) assay: A modified method[7]was adopted for the FRAP assay. The stock solutions included 300mM acetate buffer, pH 3.6, 10mM TPTZ (2, 4, 6-tripyridyl-S-triazine) solution in 40mM HCl and 20mM $FeCl_3.6H_2O$. The fresh working solution was prepared by mixing 25ml acetate buffer, 2.5ml TPTZ and

2.5ml $FeCl_3$.$6H_2O$. The temperature of the solution raised to 37°C before using. Plant extracts (0.15 ml) were allowed to react with 2.85 ml of FRAP solution for 30 min in the dark condition. Readings of the colored product (Ferrous tripyridyltriazine complex) were taken at 593nm. The standard curve was linear between 200 and 1000 µM $FeSO_4$. Results are expressed in µM (Fe (II) /g dry mass and compared with that of ascorbic acid.

Total phenol[8]

2.5ml of various concentration extracts added with 0.5 ml of Folins phenol reagent and 2ml of sodium carbonate (20%). The reaction mixture was kept in boiling water bath for 1 min. the absorbance was measured at 650 nm in a spectrophotometer.

Experimental Design

Rats were divided randomly into five groups of six animals each as follows

Group-I Animals served as normal control, treated with vehicle (0.5% carboxy methyl cellulose).1ml/kg once daily for 7 days orally.

Group-II Animals served as toxic control, diabetic agent {streptozotocin (60mg/kg,i.p.)

Group III Animals received diabetic agent {streptozotocin (60mg/kg,i.p.) + methanolic extract of leaves of *Lagerstroemia microcarpa (dose I 200mg/kg b.wt)*

Group-IVAnimals received diabetic agent {streptozotocin(60mg/kg,i.p.) + methanolic extract of leaves of *Lagerstroemia microcarpa(dose II 400mg/kg b.wt)*

Group-VAnimals received diabetic agent {streptozotocin(60mg/kg,i.p.)+ standard drug glibenclamide (10mg/kg,p.o) The extract was emulsified in 0.5% w/v aqueous solution of tween-80. Gilbenclamide (Actavis pharmaceutical, Chennai, India) was used as a standard drug. Streptozotocin was dissolved in citrate buffer (pH4.5).

Induction of non-insulin dependent diabetes mellitus (NIDDM)

NIDDM was induced in overnight fasted rats weighting 20-30g by intraperitoneal administration of streptozotocin (Sigma aldrich, Bangalore, India) solution prepared in 0.1M citrate buffer pH=4.5 at the dose of 60mg/kg body weight. Diabetes was confirmed by the determination of fasting glucose concentration on the third day post administration of streptozotocin. Blood samples were collected after 1h of administration of streptozotocin on 1st ,7th,15th and 21th day. Elevation in blood glucose level was found to be constant throughout 21 days. Serum glucose level was determined by glucometer. Rats having serum glucose level between 300-400 mg/dl were selected for further study.

Sample collection: After completing the treatment of 21 days , the rats were anesthetized by diethyl ether and sacrificed. Blood samples were collected by cardiac puncture method and intermediately by tail vein method and blood glucose levels were estimated using Glucometer.

Acute toxicity activities of leaves of *Lagerstroemia microcarpa*

The acute toxicity studies of methanolic extracts from leaves of *Lagerstroemia microcarpa*was carried as per (OECD) draft guidelines 423 adopted on 17th December 2001 received from Committee for the purpose of Control and Supervision of Experimental Animals (CPCSEA). Depending on the mortality and/or the morbidity status of the animals, an average 2-4 steps may be necessary to allow judgement on the acute toxicity of the substance/extracts. This procedure is reproducible, uses very few animals and is able to rank substances/extracts in a similar manner to the other acute toxicity testing method. The acute toxic class method is based on biometric evaluation[9] with fixed doses, adequately separated to enable a substance to be ranked for classification purpose and hazard assessment.

Change in body weight: Body weight was taken before and after experiment at the intervals of 1st ,7th, 14th and 21day of study with the help of single pan balance. The change in the body weight was noted.

Estimation of glucose: Blood glucose was measured with elegance glucometer (CT-X10, Convergent Technologies, Germany) at weekly intervals, i.e. 1st ,7th, 14 th and 21day after daily administration of extract orally.

Oral glucose tolerance test

The oral glucose tolerance test was performed in overnight fasted (18h) normal rats. Rats divided into four groups, each consisting of six rats. Glucose (2.5g/kg,p.o) was fed 0.5h after the administration of extract . Blood samples were collected by the tail- vein method just prior to the drug administration (normal fasting) and at the time intervals of 0, 30, 60 and 120m after glucose loading. Blood glucose level was measured immediately by using glucose oxidase- preoxidase reactive strips and a glucometer.

Estimation of Aspartate amino transferase(AST)/ SGOT, Alanine aminotransferase (ALT)/SGPT & Alkaline phosphatase(ALP) activity

The serum aspartate aminotransferase was estimated by the method [10,11] by using AST, ALT &ALP test kit (Span Diagnostics Ltd).

Estimation of protein: Proteins react with Folin-Ciocalteau reagent to give a complex. The colour was formed due to the reaction of alkaline copper with protein and the reduction of phosphomolybdate by tyrosine and tryptophan present in the protein. The intensity of the colour depends on the amount of these aromatic amino acids present. 0.1ml of plasma and tissue homogenate was made upto 1ml with saline, then 1ml 10% TCA was added. The mixture was centrifuged, supernatant was discarded and the precipitate was dissolved in 1ml of 0.1N sodium hydroxide. From this aliquots were taken for the estimation. 4.5ml of alkaline copper reagent was added and the contents were allowed to stand at 37ºC for 10 minutes. Then 0.5ml dilute Folin's phenol reagent was added and mixed. A series of

standards of concentration range 20-100 µg and a blank were processed as for the test. The blue colour developed was read at 620 nm after 20 minutes.

Extraction of lipids from tissues and plasma[12]

A known amount of the tissue was homogenized with 2.5 ml of ethanol-ether mixture (3:1 v/v) and digested for about two hrs at 60-65ºC and the supernatant was collected. 3 ml ethanol-ether mixture was added to the residue, digested further for a period of two hrs at 60-65ºC and the supernatant was collected. 1 ml of chloroform-methanol mixture (1:1 v/v) was then added to the residue. It was again digested for one hour at 50-55°C and the supernatant was collected. The supernatant was pooled and made up to a specified volume. The plasma was also treated similarly for the extraction of lipids. This lipid extract was finally used for the estimation of free and ester cholesterol, triglycerides, free fatty acids and phospholipids.

Estimation of cholesterol

Digitonin forms 1:1 complex with free cholesterol as cholesterol digitonide. Ester cholesterol was selectively extracted with petroleum ether (30-60°C BP). Both the ester cholesterol and free cholesterol were then subjected to Liebermann-Burchard reaction for quantitation[13]. An aliquot of the lipid extract was taken, treated with 0.5 ml of digitonin in 95% ethanol and evaporated in a waterbath. This contains ester cholesterol and digitonide of free cholesterol. About 5ml of petroleum ether was added, brought to boil, cooled and centrifuged. The supernatant was removed and used for the estimation of ester cholesterol. The precipitate of cholesterol digitonide as well as the ester cholesterol solution were evaporated at 60-80°C and treated with 6 ml of freshly prepared Liebermann-Burchard reagent. The colour developed in dark for 30 minutes and reading was taken in spectrophotometer at 620 nm against a reagent blank. 100-500 µg of working standard solution was pipetted out and treated with 0.5ml of digitonin solution and evaporated to dryness at 60-80°C. A blank was prepared by evaporating 0.5ml of digitonin solution. Blank and standards were processed as for the test. The cholesterol content was expressed as mg/g wet tissue or mg/dl plasma.

Estimation of Triglycerides: Triglycerides were estimated by the method[14]. To an aliquot of dried lipid extract, 4ml isopropanol was added, mixed well and 400 mg washed alumina was then added. This was placed in a mechanical rotor for 15 minutes and then centrifuged. To 2 ml supernatant, 0.6 ml potassium hydroxide was added and incubated at 60-70°C for 15 min, cooled and 1 ml of metaperiodate solution and 0.5 ml acetyl acetone reagent were then added. It was than mixed and incubated at 50°C for 30 minutes. A series of standards of concentration of 8-40 µg triolein were treated similarly along with a blank containing only the reagents. Cooled and read at 405 nm against a reagent blank. The triglyceride content was expressed as mg/g wet tissue or mg/dl plasma.

Statistical Analysis: Values are presented as mean ± standard deviation for groups of six animals. The results were analyzed by one way analysis of variance (ANOVA) followed by post hoc Dunnett's multiple comparison test. Differences between means were considered to be statistically significant at ($p \leq 0.05$).

3. Results and Discussion

The methanolic extract of leaves of Lagerstroemia microcarpawas subjected to screening for its phytochemical constituents. The phytochemical screening results are shown in Table 1. The methanolic extract of leaves of Lagerstroemia microcarpa containing carbohydrates, phenolic compounds, saponins, tannins, amino acids, Protein & flavonoids.

Table 1: Phytochemical analysis of methanolic extracts of leaves of *Lagerstroemia microcarpa*

S.No.	Test	Methanolic extract
I	Alkaloids	-
II	Carbohydrates and glycosides	+
III	Phytosterols	-
IV	Fixed oil and fats	-
V	Saponins	+
VI	Phenolic compounds and tannins	+
VII	Protein and Amino Acid	+
VIII	Gum and Mucilage	-
IX	Test for flavanoids	+

+ Positive,
- Negative

The methanolic extract of leaves of *Lagerstroemia microcarpa* exhibited results are shown in table 2. Maximum DPPH scavenging activity of 59.24% at 1000 µg/ml whereas for Rutin (standard) was found to be 69.83% at 1000 µg/ml. The IC_{50} of them ethanolic extract of leaves of *Lagerstroemia microcarpa* and Rutin were found to be 232 µg/ml and 480µg/ml respectively.

Table 2: Effect of methanolic extracts of *Lagerstroemia microcarpa*on DPPH assay

S.No	Concentration (µg/ml)	% of activity (±SEM*)	
		Sample (Methanolic extract)	Standard (Rutin)
1	125	47.16±0.044	18.85 ± 0.076
2	250	52.72± 0.12	22.08 ± 0.054
3	500	55.47±0.18	52.21 ± 0.022
4	1000	59.24±0.16	69.83 ± 0.014
		IC_{50} = 232«g/ml	**IC_{50} = 480«g/ml**

*All values are expressed as mean ± SEM for three determinations

The hydroxyl free radical scavenging potential shown in table 3.Maximum activity is 63.81 at 1000µg/ml for as Standard (ascorbate) was found to be 62 at 1000 µg/ml. The $IC_{50}$of the methanolic extract of *Lagerstroemia microcarpa* and standard (Ascorbate) were found to be 224 µg/ml and 410µg/ml better antioxidant is respectively.

Table 3: Antioxidant activity of methanolic extract of *Lagerstroemia microcarpa*on Hydroxy radical method

S.No	Concentration (µg/ml)	% of activity (±SEM)	
		Sample (Methanolic extract)	Standard(Ascorbate)
1	125	43.18±0.06	27.63±0.076
2	250	54.02±0.18	49.53 ±0.054
3	500	59.12±0.04	55.12±0.022
4	1000	63.81±0.08	62.00±0.014
		IC_{50}=224 «g/ml	**IC_{50}=410«g/ml**

*All values are expressed as mean ± SEM for three determinations

Maximum ferric reduced ability are shown in table 4. Maximum reduced ability 58.02 at 1000µg/ml for as Standard (Ascorbate) was found to be 98.07 at 1000 µg/ml. The IC_{50} of the methanolic extract of *Lagerstroemia microcarpa* and standard (Ascorbate) were found to be 504 µg/ml and 50µg/ml in better antioxidant is respectively.

Table 4: Reducing ability of methanolic extracts of *Lagerstroemia microcarpa*by FRAP method

S.No	Concentration (µg/ml)	% of activity (±SEM)	
		Sample (Methanolic extract)	Standard (Ascorbate)
1	125	41.14±0.16	72.04±0.014
2	250	46.06±0.14	82.05±0.034
3	500	50.12±0.06	86.04±0.026
4	1000	58.02±0.14	98.07±0.041
		IC_{50}= 504 «g/ml	**IC_{50}=50«g/ml**

*All values are expressed as mean ± SEM for three determinations

The methanolic extract of leaves of *Lagerstroemia microcarpa*was found 5.77mg/g of phenolic compound.

Table 5: The total phenolic content ofmethanolic extracts of *Lagerstroemia microcarpa*

S.No	Extract	Total Phenolic content (mg/g Catechol)	Total Phenolic content ±SEM
1	Methanolic extracts of *Lagerstroemia microcarpa*	5.54 5.86 5.92	5.77±0.08

*All values are expressed as mean ± SEM for three determinations

The results of acute toxicity study revealed that LD_{50} values of methanolic extract of *Lagerstroemia microcarpa*was high and apparently showed the safety of those extract. The treatment of rats with methanolic extract of *Lagerstroemia microcarpa*did not change the autonomic or behavioural responses among rats. The zero percent mortality for methanolic extract of *Lagerstroemia microcarpa*was found at the doses of 2000mg/kg. Based on the results, one tenth (200mg/kg) of the dose was selected as therapetuic dose of the present investigation.There were observable changes in the body weight of methanolic extract of *Lagerstroemia microcarpa*treated and STZ induced diabetic rats are shown in table 6. Treatment of diabetic rats with the methanolic extract of glibenclamide improved the weight gain compared to STZ induced diabetic rats.

Table 6: Effect of metabolic extracts of *Lagerstroemia microcarpa*on average body weight changesSTZ – induced diabetic rats

Groups	Body weight(g)			
	Day 1	**Day 7**	**Day 14**	**Day 21**
Group I	121.33±3.15^{bNS}	120.45±5.48^{b**}	120.36±5.48^{b**}	120.12±4.16^{b**}
Group II	127.22±4.16^{aNS}	126.54± 6.62^{a**}	125.14± 6.64^{a**}	124.34 ± 6.48 ^{a**}
Group III	131.43 ± 5.82$^{a*, b*}$	132.38±6. 44$^{a*, b**}$	133.48±5.54$^{a*, b**}$	134.26 ± 5.84$^{a*, b**}$
Group IV	13144± 4.48^{a*,b*}	132.62 ± 4.42$^{a*, b**}$	133.44 ± 6.34$^{a*, b**}$	134.36 ±4.58$^{a*, b*}$
Group V	128.64±5.74^{aNS,b*}	129.16 ± 6.64^{aNS,b**}	129.86 ± 7.54^{a*,b**}	130.45± 6.42$^{a*, b*}$

Values are expressed as mean ± SE (n=6 rats) P values: *<0.001, **<0.05 NS :Non significant
a →group I compared with groups II, III, IV, V. b →group II compared with groups III, IV, V.

There was a significant increase in blood glucose level in diabetic rats when compared with normal controls due to injection of STZ are shown in table 7. In the study, daily administration of the extract for 21 days led to a dose dependent fall in blood glucose levels. At the end of experiment (the 21st day) blood glucose level was (189.15 ± 0.52) and (164.34 ±0.44 mg/dL) at the doses of 200 and 400 mg/kg of methanolic extract of *Lagerstroemia microcarpa* respectively.

Table 7: Antidiabetic Effect of methanolic extracts of *Lagerstroemia microcarpa*on glucose level in STZ – induced diabetic rats

Groups	Blood glucose levels(mg/dl)			
	Day 1	**Day 7**	**Day 14**	**Day 21**
Group I	108.16±0.24^{b*}	111.46±0.34^{b**}	112.33±0.36^{b**}	113.58±0.62^{b**}
Group II	245.53±0.58^{a**}	279.90±0.72^{a**}	308.58±0.68^{a**}	369.47 ± 0.32 ^{a**}
Group III	312.33 ± 0.42$^{a*, b*}$	281.32± 0.42^{a*,b*}	262.34± 0.46^{a*,b*}	189.15 ± 0.52 ^{a*,b*}
Group IV	313.42± 0.17^{a*,b*}	265.54 ± 0.29$^{a*, b**}$	225.57 ± 0.49$^{a*, b**}$	164.34 ±0.44$^{a*, b*}$
Group V	292.44±0.24^{a*,b*}	256.44 ± 0.42^{a*,b**}	221.32 ± 0.12^{a*,b**}	128.44± 0.42$^{a*, b*}$

Values are expressed as mean ± SE (n=6 rats) *P* values: *<0.001, **<0.05 NS: Non significant
a →group I compared with groups II, III, IV, V. b → group II compared with groups III,IV, V.

In the present study the total cholesterol and triglycerides were significant increase in by 21 days treatment with STZ induced diabetic rats. After administration of methanolic extract of*Lagerstroemia microcarpa*was significant reduced in total cholesterol and triglyceride levels in 21 days in group III and IV rats are shown in table 8. The results of present study indicated that the methanolic extract of *Lagerstroemia microcarpa*possesses significant hypoglycaemic activity. It also maintained the lipid levels and body weight of rats. The hypolipidemic effect may be due to inhibition of fatty acid synthesis[15]. In normal metabolism insulin activates the enzyme lipoprotein lipase and hydrolyses triglycerides and the deficiency in insulin results in inactivation of these enzymes thereby causing hypertriglyceridemia16,17]. There was significant reduced in liver glycogen level to 257.44±6.78 (p<0.001) on day 21 in STZ treated diabetic control group II. Similarly of methanolic extract of *Lagerstroemia microcarpa* and standard drugs were significantly (p<0.001) increased the glycogen content to 466.26±6.22 and 424.24±5.46 in STZ-induced diabetic group IV and group V .

Table8: Anti diabetic Effect of methanolic extracts of *Lagerstroemia microcarpa*on STZ – induced diabetic rats

Groups	**Total Cholesterol (mg/dl)**	**Triglycerides (mg/dl)**	**Liver glycogen (mg/100g)**
Group I	102.33±0.38 b*	97.42±0.46 b*	515.18±3.42 b*
Group II	282.34±0.42 a*	169.34±0.32 a*	249.42±5.18 a*
Group III	157.64±0.46 a* b*	125.29±0.24 a* b*	486.46 ±5.24 a* b*
Group IV	118.34±0.35 a*, b*	106.42±0.44 a*, b*	466.26±6.22 a*, b*
Group V	105.22±0.32 a*, b*	95.42±0.38 a*, b*	424.24±5.46 a*, b*

Values are expressed as mean ± SE (n=6 rats) P values: *<0.001, **<0.05 NS :Non significant
a →group I compared with groups II, III, IV. b →group II compared with groups III, IV.

In the present study the SGOT and SGPT levels were significant increase in by 21 days treatment with STZ induced diabetic rats. After administration of methanolic extract of *Lagerstroemia microcarpa* was significant reduced in SGOT and SGPT levels in 21 days in group III and IV rats are shown in table 8. There was significant increased

ALP ($p<0.001$) on day 21 in STZ treated diabetic control group II). The methanolic extract of *Lagerstroemia microcarpa* and standard drugs were significantly ($p<0.001$) decreased the ALP in STZ-induced diabetic group IV and group V.

Table 9: Antidiabetic Effect of methanolic extracts of *Lagerstroemia microcarpa*on STZ – induced diabetic rats

Groups	**SGOT (IU/L)**	**SGPT (IU/L)**	**ALP(IU/L)**
Group I	15.64±0.28 b^{*}	14.18±0.12 b^{*}	50.18±0.54 b^{*}
Group II	51.62±0.32 a^{*}	30.46±0.42 a^{*}	124.42±0.68 a^{*}
Group III	29.25±0.42 $a^{*\,b*}$	19.46±0.42 $a^{*\,b*}$	68.44±0.18 $a^{*\,b*}$
Group IV	16.32±0.42 a^{*}, b^{*}	12.44±0.42 a^{*}, b^{*}	59.44±0.54 a^{*}, b^{*}
Group V	15.72±0.22 a^{*}, b^{*}	14.32±0.26 a^{*}, b^{*}	61.62±0.54 a^{*}, b^{*}

Values are expressed as mean ± SE (n=6 rats) P values: $^{*}<0.001$, $^{**}<0.05$ NS :Non significant
a →group I compared with groups II, III, IV. b →group II compared with groups III, IV.

Administration of STZ induced diabetic rats (group II) significantly reduced the levels of insulin when compared to control group of rats. The serum insulin levels were significantly increase in STZ induced diabetic rats with methanolic extract of*Lagerstroemia microcarpa*treated rats (Group III & IV) and as well as standard drug (Group V) compared with STZ induced diabetic rats (group II) are shown in table 9. The STZ induced diabetic rats were significantly decrease in the level of Total Protein (Group II) when compared to control group of rats (Group I). Treatment of rats with methanolic extract of*Lagerstroemia microcarpa*treated rats (Group III & IV) and as well as standard drug (Group V)) significantly increase the level of Total Protein compared with STZ induced diabetic rats (group II).

Table 10: Antidiabetic Effect of methanolic extracts of *Lagerstroemia microcarpa*on STZ – induced diabetic rats

Groups	**Serum Insulin(U/ml)**	**Total Protein (mg/dl)**
Group I	14.45±0.18 b^{*}	11.76±0.14 b^{*}
Group II	2.18±0.28 a^{*}	3.45±0.28 a^{*}
Group III	10.248±0.14 $a^{*\,b*}$	7.12±0.16 $a^{*\,b*}$
Group IV	16.18±0.18 a^{*}, b^{*}	8.78±0.14 a^{*}, b^{*}
Group V	16.82±0.44 a^{*}, b^{*}	9.44±0.26 a^{*}, b^{*}

Values are expressed as mean ± SE (n=6 rats), P values: $^{*}<0.001$, $^{**}<0.05$ NS: Non significant
a→group I compared with groups II, III, IV. b →group II compared with groups III, IV.

4. Conclusion

In the present study, the administration of methanolic extract of leaves of *Lagerstroemia microcarpa* shows a significant ant diabetic effects in STZ induced diabetic rats. However Further studies are needed to be isolate the active constituents of *Lagerstroemia microcarpa* and also to be evaluate the exact mechanism of action for the antidiabetic activity. These extract also showed improvement in parameters like body weight, lipid profile, and other biochemical parameters.

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