Journal of Young Pharmacists, 2024; 16(4):706-713.
https://www.jyoungpharm.org

Original Article

Antidiabetic, Antihyperlipidemic and Protective Effect of *Cupressus sempervirens* L. Leaves Extract in Streptozotocin Induced Diabetic Rats

Sunayana Vikhe[1,*], Apeksha S. Fulsundar[1], Rahul Vikhe[2]

[1]Department of Pharmacognosy, Pravara Rural College of Pharmacy, Loni, Maharashtra, INDIA.
[2]Department of Financial Management, Sir Vishveshwaraiya Institute of Technology, Sinnar, Maharashtra, INDIA.

ABSTRACT

Background: In the conventional Indian medical system, *Cupressus sempervirens* L., a member of the Cupressaceae family, is highly significant. *C. sempervirens* is a traditional medicinal plant; has been reported in the literature that the dried leaves are used as a contraceptive and to treat inflammation, diabetes, stomach pain, toothaches, and laryngitis. Hence, the present study is planned to evaluate antidiabetic, antihyperlipidemic and protective effect of *C. sempervirens* leaves extract in streptozotocin induced diabetic rats. **Materials and Methods:** The HPLC analysis of the ethanolic plant extract was followed by an *in vivo* antidiabetic activity. In order to evaluate antidiabetic measures such as body weight, urine volume, blood glucose level, total cholesterol, triglyceride, and lipid profile as antihypertensive parameters, the Streptozotocin (STZ)-induced diabetes mellitus in a rat model is being investigated for *in vivo* activity. **Results:** The presence of phenolic acid, alkaloids, flavonoids, glycosides, terpenoid, and steroids was identified by HPLC analysis of the ethanol extract. Gallic and tannic acids were identified in the phenolic components. The animals in each group having received samples were observed on days 1, 5, 10, 15, and 20, and several parameters were assessed. A significant effect is observed within the group of ethanolic extract. According to histopathological study of pancreatic and liver tissue, the damaged structure of the pancreas and liver was improved by treatment with a larger dose of ethanolic extract. **Conclusion:** Scientific evidence for their use in antidiabetic therapy is provided by the ethanolic extract, which produced a significant antidiabetic effect.

Keywords: *Cupressus sempervirens*, Antidiabetic, Antihyperlipidemic, Streptozotocin, Ethanolic extract.

Correspondence:
Dr. Sunayana Vikhe
Assistant Professor, Department of Pharmacognosy, Pravara Rural College of Pharmacy, Loni, Maharashtra, INDIA.
Email: sunainavikhe@gmail.com
ORCID: 0000-0001-5515-4005

Received: 11-06-2024;
Revised: 05-07-2024;
Accepted: 22-08-2024.

INTRODUCTION

Diabetes Mellitus (DM) is a chronic illness characterized by insufficient or inefficient insulin synthesis by the pancreas, leading to variations in the concentration of blood glucose levels.[1] Diabetes mellitus is divided into two categories: Type I diabetes (insulin dependent) and Type II diabetes (non-insulin dependent) mellitus. An autoimmune condition known as type I diabetes results in a localized inflammatory response around or within islets, which is followed by the selective death of insulin-secreting cells. While Type II diabetes is characterized by peripheral insulin resistance and reduced insulin secretion.[2] A higher risk of multiple outcomes, such as peripheral vascular disease, stroke, neuropathy, renal failure, retinopathy, blindness, and amputations, is associated with Diabetes Mellitus (DM).[3]

The fact that there is no optimal treatment for diabetes, why should we prefer the expensive, frequently associated side effects of traditional antidiabetic drugs.[4] Herbs used in Ayurveda are less expensive and have less side effects, they serve as a better alternative.[5] Several compounds found in medicinal plants possess anti-diabetic properties. Alkaloids, aromatic acids, carotenoids, coumarins, flavonoids, glycosides, organic acid, phenols and phenolics, phytosterols, protease inhibitors, saponins, steroids, tannins, terpenes, and terpenoids are the major groups of phytochemicals.[6]

Early detection of diabetes is the most effective approach improving their overall health of patients with diabetes. When given a diagnosis of diabetes, a person can maintain their health with a variety of lifestyle measures, such as regular exercise helps to keep their health in balance.[7] Plants can affect BGL in a variety of ways, such as by inhibiting the potassium channel in pancreatic beta cells, stimulating secondary messengers, adrenomimeticism, providing heavy metals that are necessary for beta cell function, and preventing the production of free radicals, which may

DOI: 10.5530/jyp.2024.16.90

contribute to the dysfunction of insulin-secreting cells in diabetes mellitus.[8] Herbal secondary metabolites may potentially lower blood glucose levels by slowing the uptake of carbohydrates.[9]

Mediterranean cypress, also known as *Cupressus sempervirens* L., is an ornamental tree of the *Cupressaceae* family. Northern America, Africa, southeastern Europe, and western Asia able to be referenced as habitats for the species.[10] The herb has been used for many years in traditional medicine, mostly to cure the flu and cough. *C. sempervirens* has been shown in studies to possess a variety of antibacterial, antiviral, insecticidal, antihyperlipidemic, cytotoxic, antioxidant, antiplatelet, hepatoprotective, and neurobiological properties.[10,11]

Dried leaves of the traditional medicinal plant *C. sempervirens* are used as a contraceptive and to treat inflammation, diabetes, toothaches, stomach pain, and laryngitis.[12] The percentages of α-amylase and α-glucosidase inhibition have been determined to assess the plant extract's potential as an antidiabetic. Increasing the tested quantity of the extract resulted in an increased inhibition of both α-amylase and α-glucosidase.[13] However, the review of the literature found no scientific evidence of the plant's leaves having an antidiabetic effect. Thus, the current study aims to investigate the antidiabetic potential of *C. sempervirens* leaf extract using Streptozotocin (STZ)-induced diabetic rats.

MATERIALS AND METHODS

Drugs and Chemicals

Streptozotocin (STZ) was purchased from Biogenuix Medsystems Pvt. Ltd., New Delhi. Metformin received from Merck, Germany. The other chemicals such as acetone, ethanol, methanol, chloroform, ethyl acetate, sodium citrate dihydrate of analytical grades were procured from PCL, India. Accu check glucometer were acquired from Roche diabetes care Inc.

Plant material and animals

The leaves of *C. sempervirens* were obtained from Sahyadri Nursery, Rahata, Maharashtra. It was authenticated by Dr. Wabale Anil Sopanrao, Head of Department of Botany, PVP College of Arts, Science, and Commerce, Pravaranagar, Maharashtra, India, via letter number PVPC/Bot/2023-24/201 dated 1/11/2023. The food and animals were purchased from Lacsmi Biofarms Pvt. Ltd., Pune. The test protocol was approved by institutional animal ethical committee (1942/PO/Re/S/17/CPCSEA/2023/03).

Preparation of extract

C. sempervirens dried and powdered 100 g of leaves were mixed together with ethanol, acetone, and chloroform to prepare extracts by a cold maceration process for 72 hr. Filter paper was used to separate the filtrate from the marc, and a rotary evaporator was used to concentrate the mixture. The extracts were then dried, and the resulting dry extract was then put into airtight bottles and refrigerated at -4°C.[14]

HPLC-DAD

Exactly 200 mg of dried extract of the *C. sempervirens* leaves was weighed and dissolved in 5 mL of acetonitrile-methanol-water (2:2:1, v/v). It was kept overnight for 12 hr at room temperature in an airtight HPLC-grade container and then put in a water bath at 55°C for 10 min. Furthermore, the sample was ultrasonicated for 10 min in order to accelerate the solid-liquid extraction and then filtered through a 0.45-um nylon filter. A freshly prepared sample of 20 uL was injected into the HPLC-DAD instrument to investigate medicinal phytoconstituents from *C. sempervirens*.

Animals

Male wistar rats (250-300 g) was used for the screening of antidiabetic activity. The rats were kept in normal housing with 22±2°C temperature, 55±10% relative humidity, and 12/12 hr light/dark cycles. They were also given a particular rodent diet and unlimited water

Acute toxicity study:

The lethal dose fifty (LD_{50}) of *C. sempervirens* extract was previously found to be 800 mg/kg bwt,[15] whereas 5%, 10% & 20% of the LD_{50} of the plant were found to be safe.[16]

Induction of diabetes

Rats were given a single IP (Intraperitoneal) injection of STZ dissolved in 0.1 M sodium citrate buffer (pH 4.5) of 55 mg/kg body weight (b.w.) dose to induce diabetes after an overnight fast. Once the animals received their STZ injection, they were given free accessibility to food and water. For 18 hr, the animals were housed in a 5% glucose solution to recover from the hypoglycemia shock. It was confirmed that diabetes had developed on the fifth day following the STZ injection by measuring blood glucose levels. The animals have been identified to have diabetes when their fasting blood glucose levels exceeded 250 mg/dL and were used for experimentation.[17]

Experimental design

The animals were divided into twelve groups of six animals each and were treated for 21 days.

Group Number	Treatment given
I	Normal Control Group [NC] group of rats that received a standard diet and had unlimited access to water.
II	Negative Control Group (STZ) [DC] group of rats that received a standard diet and had unlimited access to water.
III	Positive Control Group [PC] group of rats that received standard drug metformin (150 mg/kg b.w.)

Group Number	Treatment given
IV	The group of diabetic rats that received a dose of 40 mg/kg b.w. of CSEE.
V	The group of diabetic rats that received a dose of 80 mg/kg b.w. of CSEE.
VI	The group of diabetic rats that received a dose of 160 mg/kg b.w. of CSEE.
VII	The group of diabetic rats that received a dose of 40 mg/kg b.w. of CSAE.
VIII	The group of diabetic rats that received a dose of 80 mg/kg b.w. of CSAE.
IX	The group of diabetic rats that received a dose of 160 mg/kg b.w. of CSAE.
X	The group of diabetic rats that received a dose of 40 mg/kg b.w. of CSCE.
XI	The group of diabetic rats that received a dose of 80 mg/kg b.w. of CSCE.
XII	The group of diabetic rats that received a dose of 160 mg/kg b.w. of CSCE.

The groups mentioned above were studied for various biochemical tests employed to assess the antidiabetic activity of the samples.

Parameters study

The anti-diabetic potential of the extract was investigated by estimating various parameters such as Body Weight (BW), Urine Volume (Vu), Blood Glucose Level (BGL), Total Cholesterol (TC), Triglycerides (TG), High Density Lipoprotein (HDL), Low Density Lipoprotein (LDL), and Very Low-Density Lipoprotein (VLDL).

Histopathological studies

The rats were sacrificed at the end of the experiment by cervical dislocation. The liver and pancreas of each group were isolated, eradicated and washed using ice-cold saline solution. Pieces of both were stored in a 10% formalin solution. For histological examinations under a light microscope, paraffin portions of both tissues were pigmented with hematoxylin and eosin.[18]

Statistical analysis

Results were reported as mean±Standard Error Mean (SEM). All of the collected data were statistically analyzed using one-way ANOVA followed by Tukey's *post-hoc* test (Graph Pad Prism 10.2). Dunnett's comparison test was used to determine the statistical significance between the drug-treated groups and the negative control group ($p < 0.05$ was considered to be significant).

RESULTS

HPLC analysis

The HPLC analysis of the ethanolic plant extract revealed the presence of phenolic acid (18-20%), alkaloids (8%), flavonoids (14-16%), glycosides (40-45%), terpenoids (17-18%), and steroids (1%) (Table 1). Gallic and tannic acids were identified in the

Table 1: **Components of ethanolic extract of *Cupressus sempervirens*.**

Test for Determination	Confirmation of Components	Contribution
Phenolic acid	Detected (Gallic Acid, Tannic Acid)	18-20%
Alkaloids	Detected	8%
Flavonoids/ Polyphenols	Detected	14-16%
Glycosides	Detected	40-45%
Terpenoids	Detected	17-18%
Steroids	Detected	1%

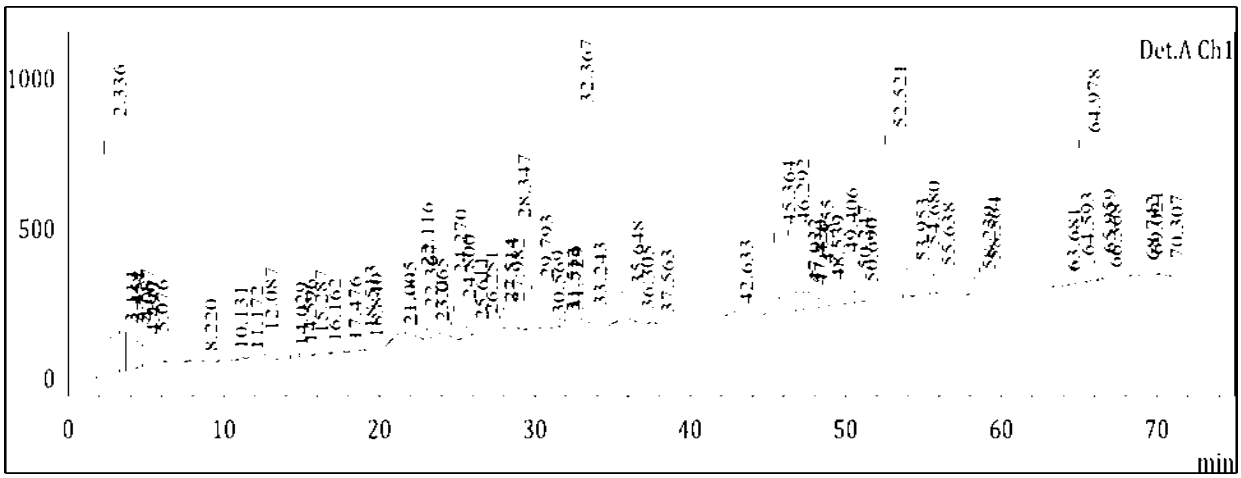

Figure 1: Ethanolic extract: HPLC chromatogram.

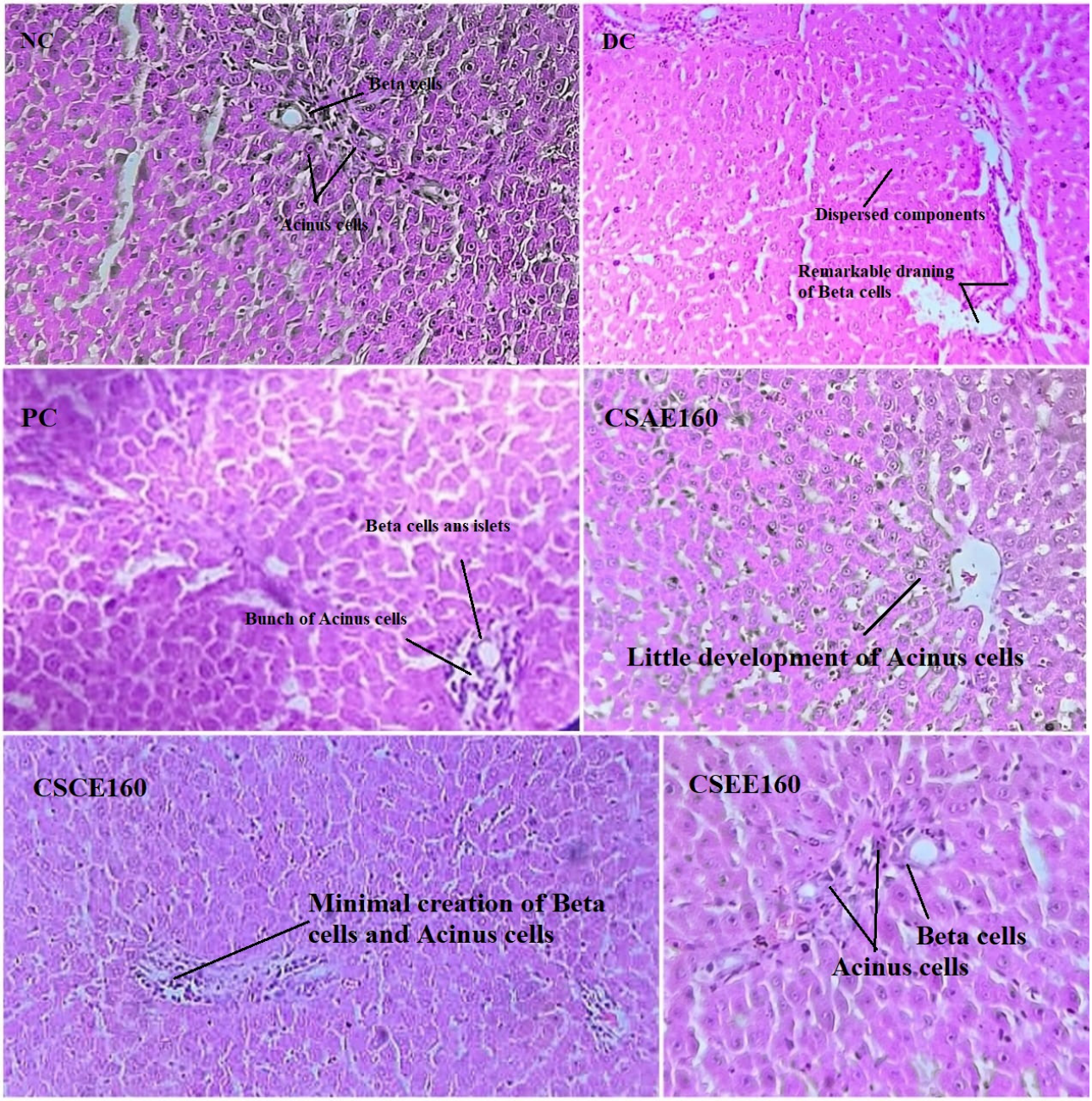

Figure 2: Histopathological findings of pancreatic tissue samples.

phenolic components. The detected phenolic compounds may differ in retention time from 3.114 to 8.220 at 210 nm (Figure 1).

DISCUSSION

Diabetes is considered to be among the most prevalent chronic illnesses in the world. There is growing public and scientific concern as oxidative stress is linked to diabetes mellitus. The STZ-induced diabetic rats are the most commonly used model to evaluate the potential of any natural or synthetic medication to prevent diabetes. STZ suppresses DNA synthesis in both bacterial and mammalian cells. STZ results in experimental diabetes, enlargement of pancreas, and islets of Langerhans beta cell degeneration.[19]

Herbs are a valuable dietary source of essential nutrients that could be evaluated for potential bioactive components for the development of new medicinal supplements. However, as much as synthetic drugs are derived from herbal origins.[20]

The HPLC analysis of the ethanolic plant extract revealed the presence of phenolic acid, alkaloids, flavonoids, glycosides, terpenoid, and steroids (Table 2). The gallic acid and tannic acid were detected in ethanol extract. In experimental rats, the presence of the compound may have an anti-diabetic effect. Previous studies have reported that the use of pharmaceutical plants to treat conditions like diabetes may be beneficial because of the high amount of different flavonoids, tannins, phenolic acid, and alkaloids.[21] Another study reports gallic acid exerts

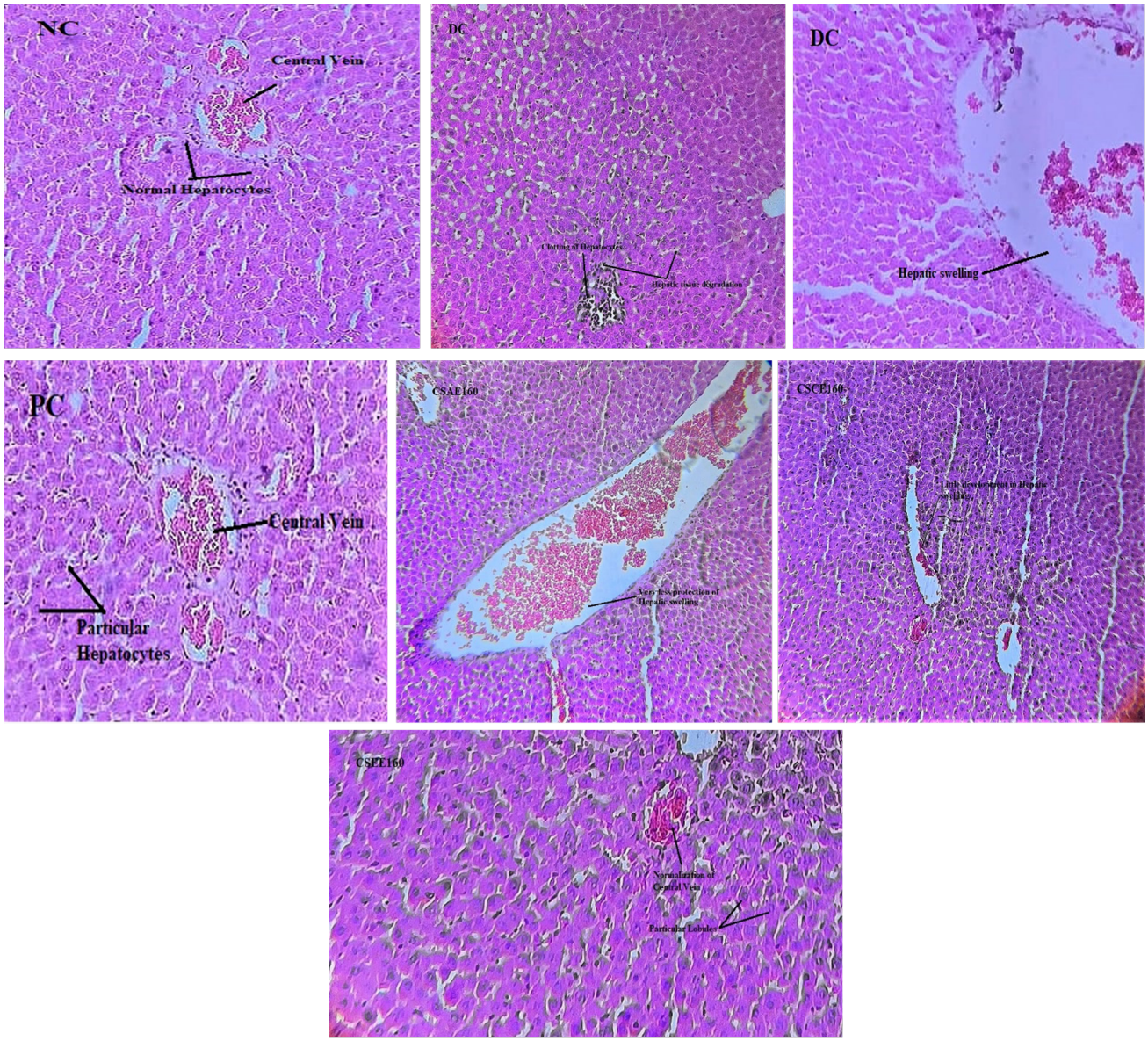

Figure 3: Histopathological findings of liver tissue samples.

Table 2: Interpretation of HPLC chromatogram of ethanolic extract .

Peak	Retention Time	Area (%)	Height (%)
1	2.336	10.696	11.350
2	3.114	2.451	1.964
3	3.451	2.016	1.926
4	3.775	0.656	1.739
5	4.087	2.538	1.895
6	4.499	1.100	1.225
7	5.076	2.575	1.236
8	8.220	0.319	0.164
9	10.131	0.373	0.231
10	11.172	0.130	0.133

Table 3: Effect of plant samples on Blood glucose level, Body weight, and Urine volume.

Parameter	Day	NC	DC	PC	CSEE 40	CSEE 80	CSEE 160	CSAE 40	CSAE 80	CSAE 160	CSCE 40	CSCE 80	CSCE 160
BW (gm)	1	263.7± 2.45	259.5 ± 4.96	264.68±2.49	266.6 ± 1.46	276.27 ± 5.37	268.61 ± 4.69	257.71 ± 5.45	269.82 ± 5.48	267.07 ± 5.16	271.43 ± 5.79	269.25 ± 5.37	271.31 ± 3.38
	5	272.24 ± 3.63	254.27 ± 6.85	273.25 ± 3.46***	267.86 ± 3.73*	275.26 ± 6.53**	270.31 ± 5.41***	248.38 ± 10.65	249.26 ± 7.07	256.49 ± 5.25	251.35 ± 2.92	256.68 ± 5.28	266.54 ± 5.02
	10	283.45 ± 5.64	226.92 ± 4.87	283.84 ± 5.53***	269.7 ± 3.93*	278.93 ± 6.03**	281.66 ± 4.26***	241.8 ± 3.9	249.51 ± 1.92	251.24 ± 1.92	249.62 ± 3.92	252.28 ± 10.7	263.37 ± 5.4
	15	284.12 ± 5.91	215.38 ± 5.45	287.68 ± 5.41***	273.86 ± 3.75*	282.1 ± 6.07**	284.16 ± 3.83***	239.24 ± 6.08	245.7 ± 2.8	251.53 ± 11.9	248.59 ± 5.3	251.25 ± 6.75	260.67 ± 4.69
	20	290.83 ± 4.71	197.25 ± 4.05	291.89 ± 8.06***	276.7 ± 3.68*	283.1 ± 6.07**	289.2 ± 3.94***	223.9 ± 8.12	232.93 ± 4.71	246.67 ± 10.3	241.45 ± 3.79	250.87 ± 6.56	256.64 ± 2.78
BGL (mg/dl)	1	80.6 ± 2.87	275.23 ± 2.89	277.58 ± 2.57	276.23 ± 3.71	276.5 ± 3.02	277.92 ± 3.78	275.86 ± 3.09	279.87 ± 3.14	284.54 ± 3.23	282.52 ± 3.24	275.53 ± 5.59	273.9 ± 3.77
	5	79.57 ± 3.45	283.03 ± 0.78	272.2 ± 2.75***	278.43 ± 0.84	278.28 ± 2.02	275.72 ± 1.07**	278.8 ± 1.43	276.63 ± 2.11	276.13 ± 1.13	275.86 ± 2.42**	275.22 ± 0.89**	273.95 ± 1.09**
	10	78.34 ± 3.72	289.85 ± 1.92	225.6 ± 3.19***	275.81 ± 2.25*	272.64 ± 3.18**	262.18 ± 3.08***	281.97 ± 1.75	279.92 ± 3.48	278.2 ± 2.16	276.18 ± 2.16	275.52 ± 1.37*	270.87 ± 2.11**
	15	77.94 ± 3.91	300.7 ± 2.98	150.84 ± 5.45***	266.76 ± 2.93*	253.3 ± 4.64**	157.51 ± 5.64***	280.69 ± 2.54	278.36 ± 2.54	277.34 ± 2.89	275.26±2.9	261.34± 9.34*	266.93±3.86*
	20	79.33 ± 3.44	328.27 ± 4.38	108.19 ± 3.95***	254.87 ± 18.72*	253.76 ± 7.02**	140.19 ± 7.88***	313.82 ± 3.21	286.21 ± 6.41	275.36 ± 18.07	273.33±18.06	273.02± 5.41	205.02±17.13**
Vu (mL/5 hr)	1	1.18 ± 0.14	7.37 ± 0.24	6.45 ± 0.39	7.22 ± 0.2	6.82 ± 0.42	6.47 ± 0.47	7.35 ± 0.41	7.17 ± 0.33	6.98 ± 0.49	8±0.42	7.2±0.55	6.9±0.45
	5	1.28 ± 0.09	8.15 ± 0.44	4.37 ± 0.44***	6.95 ± 0.36	6.83 ± 0.35	5.85 ± 0.58***	7.93 ± 0.76	7.02 ± 0.28	6.25 ± 0.56	6.42±0.43	6.6±0.18	5.88±0.64*
	10	1.3 ± 0.13	9 ± 0.69	5.15 ± 0.63***	7.17 ± 0.35	7.03 ± 0.26	5.77 ± 0.51***	6.87 ± 0.44	6.05 ± 0.72	5.88 ± 0.2**	6.83±0.26	5.67±0.62	6.43±0.49
	15	1.37 ± 0.2	9.23 ± 0.39	6.3 ± 0.15***	7.57 ± 0.44	7.15 ± 0.36	6.53 ± 0.27***	7.73 ± 0.43	7.33 ± 0.6	6.48 ± 0.64**	7.85±0.63	6.77±0.51	6.68±0.31
	20	1.28 ± 0.13	10.07 ± 0.06	5.08 ± 0.22***	7.9 ± 0.64	7.78 ± 0.44	5.22 ± 0.39***	7.8 ± 0.36	7.78 ± 0.68	5.27±0.4**	6.78±0.45	6.57±0.36	5.03±0.42***

All the values are mean ± SEM (n=6). *p<0.05, **p<0.01 and ***p<0.001 compared to control (One-way ANOVA followed by Tukey *post-hoc* test).

Table 4: Effect of plant samples on TC, TG, HDL, LDL and VLDL.

Parameter	Day	NC	DC	PC	CSEE 40	CSEE 80	CSEE 160	CSAE 40	CSAE 80	CSAE 160	CSCE 40	CSCE 80	CSCE 160
TC (mg/dl)	20	62.86 ± 0.83	174.9 ± 1.05	74.94 ± 0.6***	165.91 ± 2.11	153.96 ± 2.58**	133.8 ± 3.74***	169.05 ± 1.69	168.19 ± 1.96	163.14 ± 6.48	156.74 ± 2.71	156.47 ± 4.01	154.74 ± 2.55*
TG (mg/dl)	20	64.42 ± 0.34	189.17 ± 5.13	96.07 ± 1.05***	187.86 ± 2.86	161.99 ± 4.83**	120.28 ± 5.38***	175.28 ± 2.27	172.8 ± 4.72	170.19 ± 5.38	167.72 ± 4.51	166.3 ± 6.61	166.28 ± 2.27*
HDL (mg/dl)	20	13.8 ± 0.89	9.61 ± 0.44	13.07 ± 0.61***	11.35 ± 0.25	11.4 ± 0.3*	12.73 ± 0.14***	9.92 ± 0.44	10.25 ± 0.53	11.34 ± 0.3	10.82 ± 0.27	11.82 ± 0.2**	11.84 ± 0.2**
LDL (mg/dl)	20	24.35 ± 1.13	93.76 ± 0.79	30.46 ± 0.33***	80.59 ± 1.69	75.4 ± 2.16*	42.06 ± 3.71***	80.89 ± 3.54	77.15 ± 3.49	73.64 ± 1.44**	80.07 ± 4.38	75.04 ± 2.58*	74.98 ± 1.65*
VLDL (mg/dl)	20	14.23 ± 0.19	27.93 ± 0.67	14.74 ± 0.3***	23.99 ± 1.04*	23.86 ± 0.59**	19.44 ± 1.11***	25.4 ± 0.68	25.38 ± 1.36	24.04 ± 0.93	24.62 ± 1.09	24.03 ± 1.17	23.27 ± 0.58**

All the values are mean ± SEM (n=6). *p<0.05, **p<0.01 and ***p<0.001 compared to control (One-way ANOVA followed by Tukey *post-hoc* test).

anti-hyperglycemic effects by decreasing glucose levels and increasing insulin levels.[22]

During a 20-day period, the animals in each group having received samples were observed on days 1, 5, 10, 15, and 20, and several parameters were assessed. Anti-hyperlipidemic parameters such as Total Cholesterol, Triglyceride, High-density lipoprotein, Low-density lipoprotein, and very low-density lipoprotein and antidiabetic parameters such as Blood glucose level, Body weight, and Urine volume were investigated. Significant changes for these parameters were observed in DC animals throughout the study period.

In the present study, ethanolic extract groups showed significant decrease (p<0.001) in Blood glucose level, Urine volume, and significant increase in Body weight of animals compared to metformin-treated groups (Table 3). Compared to STZ-induced diabetic rats, rats treated with a higher dose of ethanolic extract and metformin showed an increase in High-density lipoprotein levels and a decrease in raised Total Cholesterol, Triglyceride, low-density lipoprotein, and very low-density lipoprotein levels (Table 4). Thus, it can be reported that the ethanolic extract group has a curative effect. While treatment with the ethanolic group produced significant results whereas treatment with acetone and chloroform extract were not significant.

One potential mechanism for the therapeutic effect of metformin may be that it enhances peripheral glucose absorption in the presence of insulin.[19] By suppressing alpha-glucosidase, lowering elevated oxidative stress, and repairing and regenerating beta cells, the extract is therapeutic for both hyperglycemia and cytotoxicity.

The observations of histopathological samples of pancreas and liver tissue are given in Figures 2 and 3. It was also found that the size and relative abundance of beta cells and islets were reduced in diabetic control. PC displayed common islets of Langerhans distortion in pancreatic tissue (Figure 2). CSEE160 exhibits an approximate typical structure of the islets of Langarhans. According to a histopathological analysis of pancreatic tissue, the administration of ethanolic extract altered the structure and number of islets, repaired the damage caused by STZ to the pancreatic beta cells, and preserved the integrity of the pancreatic tissue.

According to the liver tissue samples (Figure 3), the NC group was found to have normal histological features when examined under a microscope. CSEE160 showed the reducing histological alterations. Inflammation in hepatocytes was observed in diabetic control. The recovery of hepatic lobules were observed in higher dose of ethanolic extract (CSEE160).

CONCLUSION

Previously the *C. sempervirens* plant was not evaluated for diabetes or biological potential. In this project, the plant was scientifically investigated for antidiabetic activity. In the *in vivo* study, it was found that the ethanolic extract of the leaves of *C. sempervirens* was able to reduce the blood glucose level and normalize the biochemical profile as compared to negative control rats. From the HPLC analysis, we found many phytoconstituents like phenolic acids, alkaloids, flavonoid, terpenoid, and steroids in the ethanolic leaves extract of the plant, which can be responsible for this activity as these classes of compounds have already proven to have antidiabetic, hypoglycemic, or antihyperglycemic activity. However, further studies are essential for the isolation of the compound(s) of the plant, which individually or synergistically showed the activities, and it is also important to investigate the clinical study of these compounds.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

ABBREVIATIONS

DM: Diabetes mellitus; **STZ:** Streptozotocin; **HPLC DAD:** High-performance liquid chromatography with photodiode-array detection; **NC:** Normal control group; **DC:** Negative control group; **PC:** Positive control group; **CSEE:** *Cupressus sempervirens* ethanolic extract; **CSAE:** *Cupressus sempervirens* acetone extract; **CSCE:** *Cupressus sempervirens* chloroform extract; **BW:** Body weight; **Vu:** Urine volume; **BGL:** Blood glucose level; **TC:** Total cholesterol; **TG:** Triglycerides; **HDL:** High density lipoprotein; **LDL:** Low-density lipoprotein; **VLDL:** Very low-density lipoprotein.

REFERENCES

1. Mukherjee PK, Maiti K, Mukherjee K, Houghton PJ. Leads from Indian medicinal plants with hypoglycemic potentials. J Ethnopharmacol. 2006 15;106(1): 1-28. doi: 10.1016/j.jep.2006.03.021 [Epub]. PMID 16678368.
2. Arora S, Ojha SK, Vohora D. Characterisation of streptozotocin induced diabetes mellitus in Swiss Albino mice. Glob J Pharmacol. 2009;3(2):81-4.
3. Jothivel N, Ponnusamy SP, Appachi M. Antidiabetic activities of methanol leaf extract of *Costus pictus* D. Don in alloxan-induced diabetic rats. J Health Sci. 2007;53(6):655-63.
4. Dey L, Attele AS, Yuan CS. Alternative therapies for type 2 diabetes. Altern Med Rev. 2002;7(1):45-58. PMID 11896745.
5. Parasuraman S, Thing GS, Dhanaraj SA. Polyherbal formulation: concept of Ayurveda. Pharmacogn Rev. 2014;8(16):73-80. doi: 10.4103/0973-7847.134229, PMID 25125878.
6. Shaikh JR, Patil M. Qualitative tests for preliminary phytochemical screening: an overview. Int J Chem Stud. 2020;8(2):603-8. doi: 10.22271/chemi.2020.v8.i2i.8834.
7. World Health Organization. Global report on diabetes. World Health Organization; 2016.
8. Prabhakar PK, Doble M. Mechanism of action of natural products used in the treatment of diabetes mellitus. Chin J Integr Med. 2011;17(8):563-74. doi: 10.1007/s11655-011-0810-3, PMID 21826590.
9. Hashemnia M, *et al.* Blood glucose levels and pathology of organs in alloxan-induced diabetic rats treated with hydro-ethanol extracts of *Allium sativum* and *Capparis spinosa*. Afr J Pharm Pharmacol. 2012;6(21):1559-64.
10. Orhan IE, Tumen I. Potential of *Cupressus sempervirens* (Mediterranean cypress) in health. Mediterr Diet. 2015; 2014 12:639-47. doi: 10.1016/B978-0-12-407849-9.00057-9 [Epub]. PMCID PMC7149725.
11. Batiha GE, Teibo JO, Shaheen HM, Akinfe OA, Awad AA, Teibo TK, *et al.* Bioactive compounds, pharmacological actions and pharmacokinetics of *Cupressus sempervirens*. Naunyn Schmiedebergs Arch Pharmacol. 2023;396(3):389-403. doi: 10.1007/s00210-022-02326-z. PMID 36385684, PMCID PMC9898348.
12. Selim SA, Adam ME, Hassan SM, Albalawi AR. Chemical composition, antimicrobial and antibiofilm activity of the essential oil and methanol extract of the Mediterranean cypress (*Cupressus sempervirens L.*). BMC Complement Altern Med. 2014;14:179. doi: 10.1186/1472-6882-14-179, PMID 24890383.
13. Al-Rajhi AM, Bakri MM, Qanash H, Alzahrani HY, Halawani H, Algaydi MA, *et al*. Antimicrobial, Antidiabetic, Antioxidant, and Anticoagulant Activities of *Cupressus sempervirens in vitro* and *in silico*. Molecules. 2023;28(21):7402. doi: 10.3390/molecules28217402, PMID 37959821.
14. Begashaw B, Mishra B, Tsegaw A, Shewamene Z. Methanol leaves extract *Hibiscus micranthus* Linn. exhibited antibacterial and wound healing activities. BMC Complement Altern Med. 2017;17(1):337. doi: 10.1186/s12906-017-1841-x, PMID 28651570, PMCID PMC5485746.
15. Koriem KM. Lead toxicity and the protective role of *Cupressus sempervirens* seeds growing in Egypt. Rev Latinoamer Quím. 2009;37(3):230-42.
16. Koriem KM, Gad IB, Nasiry ZK. Protective effect of *Cupressus sempervirens* extract against indomethacin-induced gastric ulcer in rats. Interdiscip Toxicol. 2015;8(1):25-34. doi: 10.1515/intox-2015-0006, PMID 27486357, PMCID PMC4961923.
17. Vikhe S, Kunkulol R, Raut D. Antidiabetic and antihyperlipidemic effects of crude fractions and isolated compound from *Striga orobanchioides* Benth on streptozotocin induced diabetic rats. J Ayurveda Integr Med. 2022 26;13(3): 100618. doi: 10.1016/j.jaim.2022.100618 [Epub]. PMID 36030594, PMCID PMC9436756.
18. Histology Laboratory. Manual. Vagelos College of Physicians & Surgeons Columbia University patrice F spitalnik MD histology director. P.N. 85-6.
19. Kaur G, *et al.* Cocos nucifera in combination with metformin for modulation of diabetic symptoms in streptozotocin induced diabetic rats. J Ayurveda Integr Med. 2017. doi: 10.1016/j.jaim.2017.02.006.
20. Bahadoran Z, Mirmiran P, Azizi F. Dietary polyphenols as potential nutraceuticals in management of diabetes: a review. J Diabetes Metab Disord. 2013;12(1):43. doi: 10.1186/2251-6581-12-43, PMID 23938049, PMCID PMC7968452.
21. Kooti W, Farokhipour M, Asadzadeh Z, Ashtary-Larky D, Asadi-Samani M. The role of medicinal plants in the treatment of diabetes: a systematic review. Electron Phys. 2016;8(1):1832-42. doi: 10.19082/1832, PMID 26955456, PMCID PMC4768936.
22. Xu Y, Tang G, Zhang C, Wang N, Feng Y. Gallic acid and diabetes mellitus: its association with oxidative stress. Molecules. 2021;26(23):7115. doi: 10.3390/molecules26237115, PMID 34885698, PMCID PMC8658971.

Cite this article: Vikhe S, Fulsundar AS, Vikhe R. Antidiabetic, Antihyperlipidemic and Protective Effect of *Cupressus sempervirens* L. Leaves Extract in Streptozotocin Induced Diabetic Rats. J Young Pharm. 2024;16(4):706-13.