RESEARCH ARTICLE

Comprehensive Study of *Ehretia laevis* in Treatment of Diabetes and Related Complications

Sukeshini Lote, Mahendra Kshirsagar, Deepak Mohale, Anil Chandewar

P. Wadhwani College of Pharmacy, Yavatmal, Maharashtra, India.

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ABSTRACT

Diabetes mellitus appears to be a significant health issue globally, impacting millions of individuals and imposing an economic strain on healthcare systems all over the world. The pursuit of effective and safe alternative therapies has led researchers to investigate therapeutic plants with probable antidiabetic activity. This article highlights a comprehensive study investigating the therapeutic potential of *Ehretia laevis* in controlling of diabetes and associated complications in rats. The phytochemical investigation of *E. laevis* extracts observed phytoconstituents like flavonoids, phenolics, and triterpenoids. These phytoconstituents are known to possess antidiabetic properties. By injecting 60 mg/kg of STZ intraperitoneally once, rats were induced to develop diabetes. According to the experimental study, blood glucose levels were significantly reduced from 270 to 118 mg/dl after administering 200 and 400 mg/kg of aqueous and methanolic *E. laevis* extracts. Further evaluations on serum glutamic oxaloacetic transaminase (SGOT) and serum glutamate pyruvate transaminase (SGPT) revealed a significant reduction in their values, highlighting the control of liver complications. A lowering of serum urea and creatinine levels served to assess kidney-related complications. This comprehensive study sheds light on the potential of *E. laevis* in the treatment of diabetes and paves a path for the development of formulations, however, further mechanistic evaluation could be explored.

Keywords: *Ehretia laevis*, Diabetic mellitus, Antidiabetic, Complications.

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INTRODUCTION

A rapidly increasing common problem characterized by hyperglycemia, polyuria, glycosuria, polyphagia, and polydipsia is known as diabetes mellitus (DM). It results in an intense socioeconomic effect worldwide. Moreover, diabetes consequences are becoming more common, specifically neuropathy, nephropathy, and related cardiovascular disorders. DM, an endocrine disease, is a potential epidemic and a major cause of high medical costs for approximately 463 million people worldwide. It is predicted that 1 to 5% of Indians will develop diabetes by 2045. Natural remedies, especially herbal ones, are one of mankind's most traditional and trusted forms of therapy. People have been interested in adopting natural therapies in recent years since they have fewer potential risks and adverse effects than current pharmaceuticals.[1,2]

EL is a plant with leaves, bark, stems, seeds, and fruits all of which have therapeutic potential for various disorders. These parts have been widely used to treat several illnesses, either alone or occasionally in combination with other plants. EL has been used to manage a variety of conditions according to Ayurveda, which includes antibacterial, anti-inflammatory, injuries, wounds, astringent, fractures, antioxidant, hepatoprotective, constipation, discomfort in the teeth, cough, syphilis, gonorrhea, cachexia, and genital disorders. It has been discovered that various tribal communities employ EL leaf paste to treat both short-term and chronic inflammatory disorders.[3-5]

Botanical Description

The botanical name of this plant is *Ehretia laevis*. It is commonly known as Khanduchakka. It belongs to the division Tracheophyte and the class Magnoliopsida. It is a member of the Plantae kingdom and Boraginaceae family. This plant's genus is Ehretia and belongs to the order Boraginales. It is known as Bhairi, Chamror, Datranga, and Tamoriya in Hindi. In Marathi, it is called Ajaanvruksha or Datrang. Around 150 species in the genus *Ehretia* are cultivated mainly in India, Africa China, Nepal, Sri Lanka, Bhutan, Australia, and Vietnam. Into studies, leaves, fruits, and bark of the EL plant were rich sources of minerals such calcium, magnesium, potassium, sodium, copper, ferrous, zinc, phosphorous, ammonia, manganese, silicon, proteins, and lipids.[6-7] Additionally, the plant comprises pentacyclic triterpenoids, flavonoids, phenolic acids, alkaloids, tannins, amino acids, carbohydrates, alcohols, ascorbic acid,

**Author for Correspondence: sblote10@gmail.com*

hydrocarbons, and benzoquinones. Pentacyclic triterpenes found in EL have recently gained a lot of attention due to their diverse biological functions. Pentacyclic triterpenoids obtained from chloroform, petroleum ether, and methanolic extracts of EL are lupane, oleanane, ursane, ursolic acid, betulinic acid, α and β-amyrin, botulin, lupeol, β-sitosterol, bauerenol, and its acetate form. These substances exhibit a range of pharmacological behaviors and are typically not very harmful. Thus, the scientific community has identified these triterpenes as the most promising and likely candidates for the development of newer multi-targeting bioactive medications.[8-10]

MATERIAL AND METHOD

Animals

Healthy Sprague dawley rats, about eight weeks old, weighing between 180 to 250 g, were employed for current research. The rats were kept in a cage made of polypropylene wire mesh with husk bedding. They were kept in a controlled environmental condition of light like 10 hours light, and 14 hours dark, 22 ± 3°C temperature, and humidity. They were fed a conventional pellet diet and water available all time and can drink when required.[11] The Institutional Animal Ethics Committee (IAEC) has granted approval to the protocol. The animal study was conducted as per CPCSEA regulations under research protocol no. 650/PO/Re/S/2002/CPCSEA/2022/08.

Chemicals

Ambika Diagnostics provided the diagnostic kits (SGOT, SGPT, urea, creatinine, and blood glucose estimate). Cipla Pvt Ltd., Mumbai, graciously donated glibenclamide, and M. P. Biomedical provided streptozotocin. Other chemicals required were of laboratory grade. Biochemistry analyzer AD-100 (Ambica Diagnostics, Parbhani, India), cooling centrifuge (Remi Elektrotecnik Ltd., Vasai, India), micropipette (10–100 µL), anesthetic chamber, weighing machine are utilized in the current work.[12]

Plant Material

The EL leaves were gathered from several locations around Maharashtra. Plant was recognized and confirmed by Mrs. A. M. Gaharwar, Krishi Vigyan Kendra, Yavatmal (Ref No. (Ref No. VNCABT/Ytl/Hort/1105/2022). EL leaves dried in shade before they were ground into a coarse powder. This powder was utilized for extraction and kept in an airtight container. The dried powder was then subjected to methanol extraction by maceration and infrequently stirred at steady intervals. Then it was filtered and dried using a water bath. The remaining plant material is then macerated with water. The extract was screened for the presence of preliminary phytochemicals like flavonoids, triterpenes, and other plant metabolites.[13]

Experimental Design

There were seven groups of animals altogether, each group having six animals (n = 6). Animals in group I are the control group. The negative (diabetic) control group, consisting of six animals, was given a dose of STZ at a rate of 60 mg/kg. Group III animals were given 0.5 mg/kg of the antidiabetic medication glibenclamide orally once day after the diagnosis of diabetes was confirmed. Methanolic extract of *E. laevis* (MEL) was given orally to diabetic rats in groups IV and V at 200 and 400 mg/kg, respectively. An aqueous extract of *E. laevis* (WEL) was given orally to animals in group VI at a dosage of 200 mg/kg, whereas rats in group VII were given a dose of 400 mg/kg. After 72 hours of receiving single i.p. (intraperitoneal) injection of 60 mg/kg STZ, rats develop hyperglycemia.[14-15]

Assessment of Biochemical Parameters

The retro-orbital plexus piercing method was used to collect blood samples. An electronic biochemistry analyzer and commercial diagnostic kits were employed. Following the manufacturer-provided instructions, the process was executed. In order to assess the efficacy of the antidiabetic medication, the blood glucose level was monitored. Impact on renal functions related to diabetes was evaluated by measuring serum urea and creatinine levels. The levels of SGOT and SGPT were assessed to examine the impact of EL extracts on liver complications resulting from diabetes.[16,17]

RESULT

Qualitative screening of MEL and WEL to determine the presence of phytoconstituents is provided in Table 1. The testing indicates the occurrence of alkaloids, carbohydrates, flavonoids, proteins, steroids, and tannins, whereas glycosides were absent.

Figure 1 shows that on day 3, all rats studied had expressively higher BGL ($p < 0.01$) compared to the control group (I). Rats were given 200 mg/kg doses of MEL and WEL after the onset of diabetes had been confirmed. There was a substantial reduction ($p < 0.05$) in BGL in groups that were treated. Just as the negative control group (II) on 14 and 28 days, rats given 400 mg/kg of MEL and WEL exhibited a significant decrease ($p < 0.01$) in BGL.

Figure 2 shows the effect of MEL and WEL on serum urea levels in diabetic rats. On the 28th day, associated with the normal control group, the negative control group presented a substantial growth ($p < 0.01$) in urea levels. On the 28th day after taking 200 and 400 mg/kg of MEL and WEL, a notable reduction ($p < 0.01$) in serum urea level was noted.

Effect of MEL and WEL on blood creatinine level in diabetic rats is shown in Figure 3. The creatinine level in the negative control group was expressively higher ($p < 0.01$) on 28th day compared to normal control group. On 28th day, after administering both plant extracts at doses of 400 and 200 mg/kg, there was a notable decrease ($p < 0.01$) in the diabetic control group's serum creatinine level.

In the negative control group of rats, SGOT level increased significantly ($p < 0.01$) on 28th day. As shown in Figure 4, SGOT level was expressively lower ($p < 0.01$) after treatment with MEL and WEL associated to the diabetic control group on 28th day.

On the 28th day, the SGPT level in the diabetic control group of rats increased expressively ($p < 0.01$). On 28th day,

Table 1: Phytochemical investigation of methanolic and aqueous extract of *E. laevis* (EL)

Phytoconstituents	*Test*	*MEL*	*WEL*
Alkaloids	Dragondorff's	+	+
	Mayer's	+	+
Glycosides	Keller Kiliani	-	-
	Molisch	+	+
Carbohydrates	Benedict	+	+
	Fehling	-	-
Flavonoids	Ferric chloride	+	+
	Lead acetate	+	+
Steroids	Liebermann Burchard	+	+
	Salkowaski	+	+
	Xanthoprotic	+	+
Proteins	Biuret	+	-
	Lead acetate	+	+
Tannins	Lead acetate	+	-
	Gelatin	+	+

+ Present - Absent

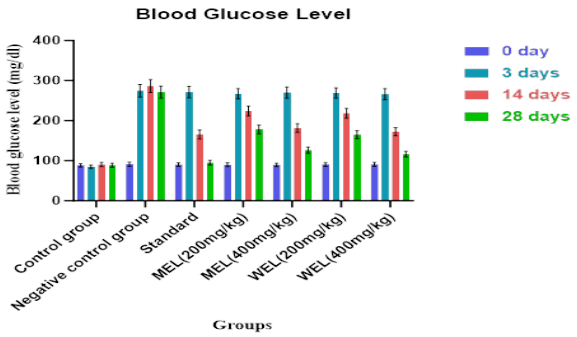

Figure 1: Effect of MEL and WEL extract on blood glucose levels

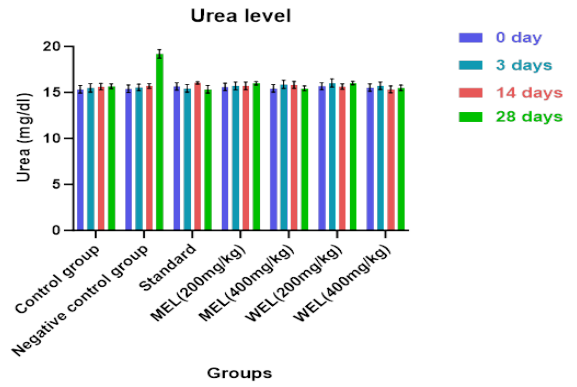

Figure 2: Effect of MEL and WEL extract on serum urea levels in diabetic rats induced by STZ

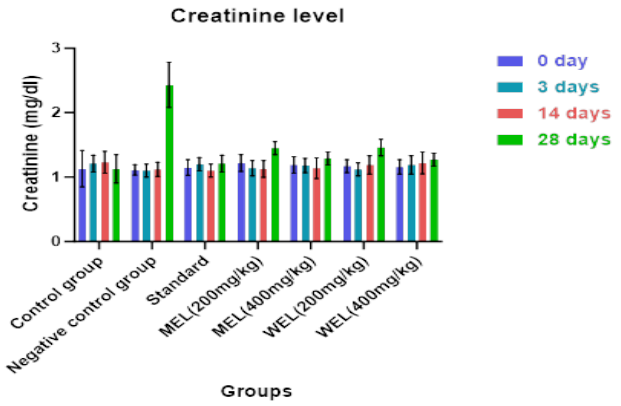

Figure 3: Effect of MEL and WEL extract on serum creatinine levels in diabetic rats induced by STZ

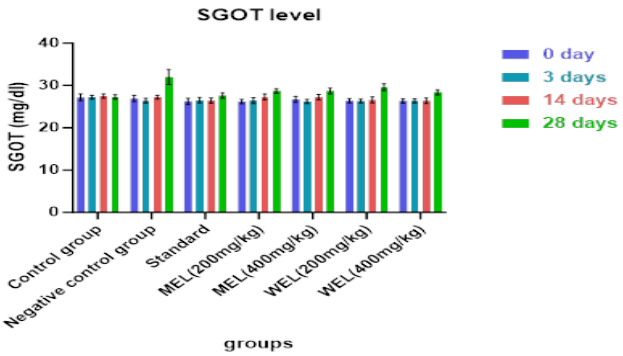

Figure 4: Effect of MEL and WEL extract on serum SGOT levels

the treated group showed expressively lower SGPT level (p <0.01) compared to the diabetic control group after oral administration of 400 and 200 mg/kg of MEL and WEL, as shown in Figure 5.

DISCUSSION

Administering STZ at dosage of 60 mg/kg has the ability to induce diabetes in rats. In Sprague dawley rats, STZ is administered i.p., resulting in toxicity to the pancreatic β-cells. Scientists have developed various animal models to produce diabetic diseases.[18] The present research investigation involved the selection of Sprague dawley rats to induce diabetes. Two crucial factors in the development of diabetes are the decrease in insulin sensitivity and malfunctioning of pancreatic β-cells.[19] One method to decrease insulin sensitivity is by adjusting the diet to include foods that are high in fat. Additionally, the use of streptozotocin can cause damage to pancreatic cells.[20] The methanolic and aqueous extracts of EL, given by 200 and 400 mg/kg doses, had a notable anti-hyperglycemic effect. Both extracts of EL leaves exhibited a significant reduction in BGL of diabetic rats. Current study indicates efficacy of extracts in treating diabetes mellitus.[21] It offers empirical evidence supporting the utilization of EL in the treatment of diabetes. The experimental diabetic rats exhibited a notable

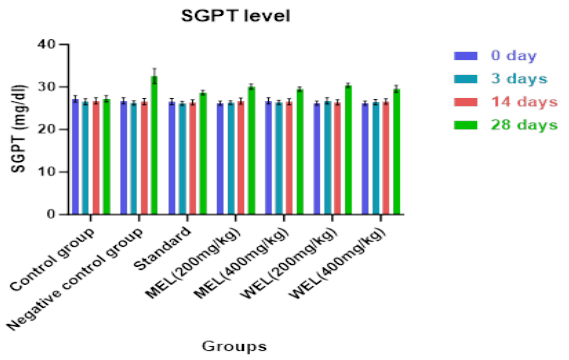

Figure 5: Effect of MEL and WEL extract on serum SGPT levels

increase in their BGL as a result of reduced insulin sensitivity and damage to the β-cells of pancreas.[22] Administration of methanolic and aqueous EL extracts effectively produces the hypoglycemic effect in diabetic rats. The BGL in all treated groups shows a major reduction ($p < 0.01$) associated to STZ group (negative control). A significant decrease in SGOT and SGPT values compared to negative control group serve as indicators of normal liver function. The decrease in treated groups' serum creatinine and urea levels indicates avoidance of diabetes-associated renal damage.[23-25]

CONCLUSION

The methanolic and aqueous extracts of *E. laevis* have a significant antidiabetic effect. This effect is due to the presence of phytochemicals like flavonoids. The mechanism of producing an antidiabetic effect can be further evaluated.

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