

# PHYTOCHEMICAL INVESTIGATION AND ANTIDIABETIC ACTIVITY OF HERBAL FORMULATION OF *OUGEINIA OOJEINENSIS* PLANT EXTRACTS

## ABSTRACT

The aim of this research to evaluate antidiabetic potential of prepared herbal formulation of *Ougeinia oojeinensis* plant extracts, presented in this thesis reveals the excellent potential of phospholipids complex based drug delivery system for improving the *in-vivo* antidiabetic activity of plant extract. There is some relation between phospholipids complex and various physicochemical attributes such as size and entrapment efficiency. The significant antidiabetic effect can be achieved by optimized phospholipids complex formulation at a lower dose (100 mg/kg and 200 mg/kg) in comparison to hydroalcoholic extract (200 mg/kg and 400 mg/kg). The extracts also showed improvement in lipid profile and body weight at a lower dose than hydroalcoholic extract. So it can be concluded that phospholipids complex formulation improved the dose efficacy ratio. Optimized phospholipids complex formulation of *Ougeinia oojeinensis* extract also inhibited the histopathological changes of the pancreas and kidney in streptozotocin-nicotinamide induced diabetes. Thus, the present study demonstrated that *Ougeinia oojeinensis* extract could be useful in management of type-1 and type-2 diabetes associated with abnormalities in lipid profiles.

**Keywords:** *Ougeinia oojeinensis*, Antidiabetic, Herbal Formulation, Phospholipids complexes

## INTRODUCTION

Diabetes mellitus is a group of diseases characterized by high levels of blood glucose resulting from defects in insulin production, insulin action, or both, altered metabolism of lipids, carbohydrates and proteins and an increased risk of complications of vascular diseases<sup>1</sup>. It is the most common endocrine disorder and can be associated with serious complications and premature death.

Knowledge of diabetes mellitus dates back to many centuries. Eber's Egyptian Papyrus (1500 BC) described it as an illness associated with passage of much urine. A renowned Greek physician Arataeus of Cappodica coined the name diabetes (a siphon which described it as melting down of flesh and limbs into urine [1-4].

Scholars in China, Japan and India during the 3rd and 6th centuries AD wrote of a condition with polyurea in which urine was 'sweet and sticky'. Although it has been for centuries, it was until 1674, when Willis added the observation 'as it imbued with honey and sugar'. The name diabetes mellitus (mellitus- honey) was thus established. A century after Willis, Dobson demonstrated that the sweetness indeed was due to sugar. From the time of the earliest recorded history of diabetes, progress in understanding came slowly until the middle of the 19th century. An association was established with a disturbance in the beta cells, clustered as tiny islets of tissue in the exocrine pancreas. Brockman first noticed the presence of these islets in fish during the 19th century. Soon after, the german scientists, Mering and Minkowski, found that surgical removal of pancreas produced diabetes in dog. The major breakthrough of discovery of insulin was by Fredrick Banting and Charles Best in 1921. By 1952, various purified modified versions of insulin were available and by 1982, recombinant DNA insulin became available. Thus diabetes, which was synonymous with an inevitably fatal disease, became a more benign controllable disease [5-8].



**Fig. 1: *Ougeinia oojeinensis***

*Ougeinia oojeinensis* belongs to the family Fabaceae. It is an herb found all around the world, and all parts of India mostly in the outer Himalayas and sub-Himalayan tracts from Jammu to Bhutan. Commonly it is known as Tinsa, Sandan, Panjan (Sharma, 2001) (Singh, 2002). The extract of the whole plant *O. oojeinensis* shows anti-inflammatory and analgesic, hepatoprotective, antioxidant, anthelmintic, hypoglycemic, antidiabetic and wound healing activity. Phytochemical investigated on *O. oojeinensis* have reported the presence of lupeol, hydroxlupeol, betulin and isoflavanones such as dalbergioidin, homoferreirin and ougenin. The bark and leaves are used in the treatment of jaundice, diarrhoea, dysentery, urorrhagia, diabetes, verminosis, leprosy, leucoderma, haemorrhages, fevers, ulcers etc. Systemic literature survey is

the main basis for the planning of any scientific work and due to the same reasons here the review of literature regarding the *Ougeinia oojeinensis* (Roxb) Hochr, has been done under various heading like pharmacognostical review, phytochemical review, pharmacological review and ethanomedical information. Botanical classification of *Ougeinia oojeinensis*: Arrangement of plant into groups and subgroups is commonly spoken as classification. The foundation of taxonomical is mainly laid down by International code of botanical nomenclature, binomial nomenclature mainly indicates in designating a plant in terms of it, genus name and species names. A large number of plant families have certain distinguishing characteristics that permits crude drug from these families to be studied at one time. It is scientific way of naming, describing and arranging the plants in an orderly manner [8-13].

## **Material & Method**

### **Collection and authentication of Plant material**

The plant *Ougeinia oojeinensis* leaves, were collected and authenticated. The plant material were dried under shade, pulverised and kept in air tight container.

### **Phytochemical Screening**

#### **Qualitative Phytochemical Screening**

Preliminary phytochemical screening of the extracts of *Ougeinia oojeinensis* was carried out. The crude extracts were screened for the presence or absence of secondary metabolites such as alkaloids, steroidal compounds, phenolic compounds, flavonoids, saponins and tannins by using standard procedures [3-4].

#### **Test for alkaloid [3]**

The chemical test were performed from neutral or slightly acidic solution of drug; following type of chemical test given by alkaloids are-

#### **Dragendorff's test:**

Dragendorff's reagent (Potassium bismuth iodide) was added to the plant extract. Appearance of orange red coloured precipitate, confirmed the presence of alkaloids.

#### **Mayer's test:**

Adequate amount of plant extract was taken and few drops of Mayer's reagent (Potassium mercuric iodide) were added. Appearance of creamy-white coloured precipitate, confirmed the presence of alkaloids.

**Hager's test:**

Adequate amount of plant extract was taken and few drops of Hager's reagent (Saturated aq. solution of picric acid) were added. Appearance of crystalline yellow coloured precipitate, confirmed the presence of alkaloids.

**Wagner's test:**

Few drops of Wagner's reagent (Dilute iodine solution) were added to the plant extract. Appearance of reddish-brown coloured precipitate, confirmed the presence of alkaloids.

**Tannic Acid test:**

Few drops of tannic acid solution were added to the plant extract. Appearance of buff coloured precipitate, proved the presence of alkaloids.

**Test for carbohydrates [4]****Charring test:**

The plant extract was taken in a test-tube and heating was done in presence of conc.  $H_2SO_4$  produces charring with smell like burning sugar confirmed the presence of carbohydrate.

**Molish test:**

The plant extract was taken in a test tube and few drops of Molish reagent ( $\alpha$ -naphthol) and conc.  $H_2SO_4$  was added from sidewall of test tube. Appearance of purple coloured ring at junction, confirmed the presence of carbohydrates.

**Iodine test:**

To the plant extract few drops of iodine solution were added. Appearance of blue colour, which disappeared on heating and reappeared on cooling, revealed the presence of starch. This test is specifically for polysaccharides.

**Barfoed test:**

To the plant extract 2 ml of Barfoed reagent (Cupric acetate, acetic acid and water) was added and boiled. Appearance of brick red precipitate in 5 min, revealed the presence of monosaccharide while in 7 min, revealed the presence of disaccharide. It is used to make difference between monosaccharides and disaccharides.

**Fehling solution test:**

The test is basically used for reducing sugars and composed of two solutions, which are mixed in situ. Fehling solution A composed of 0.5% of copper sulphate whereas Fehling solution B composed of sodium potassium tartarate. Equal volumes of Fehling A and Fehling B solutions

were mixed (1 ml each) and 2 ml of plant extract was added followed by boiling for 5-10 min on water bath. Appearance of reddish brown coloured precipitate due to formation of cuprous oxide, confirmed the presence of reducing sugar.

**Benedict's test:**

To the plant extract 1 ml of Benedict's solution (Copper sulphate and sodium hydroxide) was added and heated almost too boiling, appearance of green, yellow, orange, red or brown colour in order of increasing concentration of simple sugar in the test solution, due to formation of cuprous oxide, showed the presence of reducing sugars.

**Test for protein and amino acids [3]:**

**Biuret test:**

Few drops of Biuret reagent (Potassium hydroxide, copper sulphate and sodium potassium tartrate) were added to the plant extract. Appearance of violet or pink colour, showed the presence of proteins.

**Millons test:**

Few drops of Millons reagent (solution of mercuric and mercurous ions in nitric and nitrous acids) were added to the plant extract. White precipitate appears that turns red after heating on boiling water bath, showed the presence of proteins.

**Ninhydrin test:**

Few drops of 5% Ninhydrin solution were added to the plant extract and heated in boiling water bath for 10 min. Appearance of purple or bluish colour showed the presence of proteins.

**Lead sulphide test:**

2 ml of 10% NaOH solution was added to the plant extract followed by few drops of lead acetate solution. The solution was shaken and boiled on water bath for few minutes. Appearance of black precipitate in presence of sulfur containing amino acids, showed the presence of proteins.

**Xanthoproteic test:**

Adequate amount of plant extract was taken and 2 ml of dil.  $\text{HNO}_3$  was added. Appearance of yellow precipitate, showed the presence of aromatic ring containing amino acid like tyrosine, tryptophan etc.

**Test for steroids [3]:**

**Liebermann burchard test:**

Plant extract was firstly extracted with chloroform and then few drops of acetic anhydride were added followed by conc.  $\text{H}_2\text{SO}_4$  from side wall of test tube. A violet to blue coloured ring appeared at the junction of two liquid that showed the presence of steroid entity.

**Salkovaski test:**

Plant extract was firstly extracted with chloroform and then conc.  $\text{H}_2\text{SO}_4$  was added from sidewall of test tube. Appearance of yellow coloured ring at the junction of two liquid, which turns red after 2 min, confirmed the presence of steroid.

**Trichloro acetic acid test:**

Adequate amount of plant extract was taken and saturated solution of trichloro acetic acid was added. Colored precipitate appeared that proved the presence of steroid moiety.

**Test for glycosides [3]:**

**Borntrager's test:**

To the appropriate amount of plant extract dil.  $\text{HCl}$  was added, boiled and filtered. Equal volume of chloroform was added to the filtrate and shaken well. Chloroform layer was separated and ammonia was added. Appearance of red colour in ammonical layer, showed the presence of anthraquinone.

**Modified Borntrager's test:**

An appropriate amount of plant extract was taken and dil.  $\text{HCl}$  was added followed by 5 ml  $\text{FeCl}_3$ . Boiling was done for 10 min on water bath, cooled and filtered, filtrate was extracted with  $\text{CCl}_4$  or benzene and equal volume of ammonia solution was added. Appearance of pink to red colour, revealed the presence of anthraquinone moiety.

**Keller Killiani test:**

To the adequate amount of plant extract equal volume of water and 0.5 ml of strong lead acetate solution was added, shaken and filtered. Filtrate was extracted with equal volume of chloroform. Chloroform extract was evaporated to dryness and residue was dissolved in 3 ml of glacial acetic acid followed by addition of few drops of  $\text{FeCl}_3$  solution. The resultant solution was transferred to a test tube containing 2 ml of conc.  $\text{H}_2\text{SO}_4$ . Reddish brown coloured layer appeared which turns bluish green after standing, confirmed the presence of digitoxose.

**Legal test:**

An appropriate amount of plant extract was taken and equal volume of water and lead acetate solution was added, shaken and filtered. Filtrate was extracted with equal volume of chloroform

and the chloroform extract was evaporated to dryness. The residue was dissolved in 2 ml of pyridine and 2 ml of sodium nitropruside was added followed by addition of NaOH solution to make alkaline. Appearance of pink colour, showed the presence of glycosides or aglycone moiety.

**Baljeet test:**

Sodium picrate solution was added to the plant extract. Yellow to orange colour appears that revealed the presence of aglycones or glycosides.

**Test for saponins:**

**Haemolysis test:**

Few drops of plant extract were mixed with a drop of blood on slide. Appearance of ruptured RBC's, revealed the presence of saponins.

**Foam test:**

10-20 ml of water was added to the plant extract and shaking was done for few minutes, Appearance of froth which persisted for 60-120 s, confirmed the presence of saponins.

**Test for flavonoids:**

**Ammonia test:**

A piece of filter paper was dipped in the plant extract then exposed to ammonia vapor. Appearance of yellow spot on filter paper, showed the presence of flavonoids.

**Shinoda test**

(a) Magnesium turning and dil. HCl were added to the plant extract. Appearance of red colour, showed the presence of flavonoids.

(b) Zn turning and dil. HCl were added to plant extract. Deep red to magenta colour appeared that revealed the presence of dihydro flavonoids.

**Vanillin-hydrochloric acid test:**

Vanillin hydrochloric acid was added to plant extract. Appearance of pink colour, revealed the presence of flavonoids.

**Test for phenolic compounds [4]:**

**5% Ferric chloride solution:** An appropriate amount of plant extract was taken and 5% FeCl<sub>3</sub> was added. Appearance of deep blue-black colour, confirmed the presence of phenolic compounds.

**Lead acetate solution:** Lead acetate solution was added to plant extract and appearance of white colored ppt., confirmed the presence of phenolic compounds.

**Gelatin solution:** Gelatin solution was added to the plant extract. Appearance of white ppt., revealed the presence of phenolic compounds.

**Potassium dichromate:** Appearance of red ppt., revealed the presence of phenolic compounds.

**Dilute iodine solution:** Appearance of transient red colour, revealed the presence of phenolic compounds.

**Dilute nitric acid:** Appearance of reddish to yellow colour, revealed the presence of phenolic compounds.

**Acetic acid solution:** Appearance of red colour solution, revealed the presence of phenolic compounds.

**Dil. potassium permanganate solution:** Appearance of de coloration, revealed the presence of phenolic compounds.

**Test for fixed oils [5]:**

**Spot test:**

Small quantity of plant extract was taken and pressed between two filter papers. Oil stains on the paper, showed the presence of fixed oil.

**Saponification test:**

Few drops of 0.5 N alc. KOH and phenolphthalein were added to the plant extract. Mixture was heated on a water bath for 1-2 h. Soap formation or partial neutralisation takes place that confirmed the presence of fixed oils.

**Preparation of Phospholipids complex**

Phospholipid complex of plant extract of 1:1 molar ratio trial batches were prepared by using different methods i.e antisolvent precipitation technique, rotary evaporation technique and solvent evaporation technique [14-15].

**Antisolvent precipitation technique**

The particular amounts of plant extract and soya lecithin were dissolved in methanol, while cholesterol was dissolved in DCM. The mixture was refluxed at temperature not exceeding 60°C for 2 h to obtain a concentrate. An anti-solvent i.e Hexane was added deliberately with stirring to get the precipitate which was sifted and collected and stored in vacuum desiccators overnight. Powdered complex was placed in amber colored glass bottle and stored in refrigerator.

### ***In-vivo* anti diabetic activity**

The *in vivo* anti-diabetic activity was evaluated by the method previously reported in the literature [31-32]. The animals were maintained under normal laboratory condition & kept in standard polypropylene cages at room temperature of  $30 \pm 2^\circ\text{C}$  and 60–65% relative humidity and provided with standard diet & water ad libitum. The experimental protocol was approved by institutional Animal Ethical Committee & a written permission from in house ethical committee has been taken to carry out and complete this study.

#### **Oral glucose tolerance test (OGTT)**

Glucose tolerance checks how well our body's cells ready to absorb glucose load that's the main source of energy. The oral glucose resilience test (OGTT) is essential for the portrayal of metabolic disorder, the regular progression from prediabetes to Type 2 diabetes, and portrayal of the metabolic activities of cardiovascular and metabolic medications.

Different doses of plant extracts will be administered 60 min prior to oral glucose load (2.0 g/kg). Animals were randomly assigned into following groups of six animals each.

**Group I:** Control received Glucose (2g/kg)

**Group II:** received Glucose + Hydroalcoholic extract (200mg/kg)

**Group III:** received Glucose + Hydroalcoholic extract (400mg/kg)

**Group IV:** received Glucose + Phospholipids complex of Hydroalcoholic extract (100mg/kg)

**Group V:** received Glucose + Phospholipids complex of Hydroalcoholic extract (200mg/kg)

**Group VI:** received Glucose + Glibenclamide (10 mg/kg)

The blood samples will be collected from each group just before glucose administration (0 min) and at 30, 60 and 120 min after glucose administration. Blood glucose levels will be estimated using Glucometer.

#### **Streptozotocin- Nicotinamide induced diabetic model**

Streptozotocin with the chemical name 2-Deoxy-2-[[[(methylnitrosoamino) - carbonyl] amino]-D-glucopyranose, is a naturally occurring chemical that is particularly toxic to the insulin-producing  $\beta$ -cells of the pancreas in mammals [33]. It was originally identified in the late 1950s as an antibiotic [34].

#### **Experimental Protocol**

Diabetic animals were randomly assigned into following groups of six animals each.

**Group I:** Diabetic control received vehicle (Normal saline)

**Group II:** Diabetic animals received Hydroalcoholic extract (200mg/kg)

**Group III:** Diabetic animals received Hydroalcoholic extract (400mg/kg)

**Group IV:** Diabetic animals received Phospholipids complex of Hydroalcoholic extract (100mg/kg)

**Group V:** Diabetic animals received Phospholipids complex of Hydroalcoholic extract (200mg/kg)

**Group VI:** Diabetic animals received Glibenclamide (10 mg/kg)

### Treatment

Experiments were performed in rats that had been fasted overnight (deprived of food for at least 12 h but allowed free access to water). The drug solution and vehicle were introduced orally once daily for 21 days. The effect of vehicle, extract and standard drug on blood glucose, body weight was determined in fasted animals at 0, 7, 14, 21 day after oral administration. Blood sample was collected by retro-orbital plexus and cardiac puncture of all the animals at 21<sup>st</sup> day under mild anesthesia and biochemical parameters were estimated using the diagnostic kit (ERBA Diagnostic Mannheim, Germany) in Auto-analyzer.

## RESULT & DISCUSSION

### Qualitative phytochemical screening

Table 1, shows the results of qualitative phytochemical screening.

**Table 1: Results of qualitative phytochemical screening of *Ougeinia oojeinensis***

S.NO.	Class of compound		HECE
1.	Carbohydrates	Reducing sugars	+ve
		Monosaccharides	+ve
		Pentose sugar	+ve
		Hexose sugar	+ve
		Molish test	+ve
2.	Glycosides	Keller killiani	-ve
		Legal test	-ve
3.	Proteins	Biuret test	+ve

		Million's test	+ve
4.	<b>Steroids</b>	Salkowski test Liebermann-Burchard test	-ve -ve
5.	<b>Tannin</b>	Extract + Iodine solution Extract + Acetic acid Extract + 5% ferric chloride Lead acetate test	+ve +ve +ve +ve
6.	<b>Flavonoids</b>	Extract + Lead acetate Shinoda test Extract + NaOH	+ve +ve +ve
7.	<b>Amino acids</b>	Ninhydrin test	+ve
8.	<b>Alkaloids</b>	Dragendorff 's test Mayer 's test Hager 's test Wagner 's test	+ve +ve +ve +ve
9.	<b>Saponins</b>	Foam test	+ve

### ***In vivo* anti diabetic activity**

#### **Oral glucose tolerance test (OGTT)**

##### **OGTT of optimized formulation of *Ougeinia oojeinensis***

Within 30 min from starting of the glucose tolerance test, blood glucose concentration almost more than doubled from its initial level of control. This hyperglycemia was maintained until 60 min and then began to decrease (Table 2). *Ougeinia oojeinensis* significantly prevented the increase in blood glucose levels after 60 min. of glucose administration at the doses of 200 and 400 mg/kg. Phospholipid complex of *Ougeinia oojeinensis* prevented the increase in blood glucose levels after 60 min. Glibenclamide also blocked the increase in blood glucose levels after 30 min.

**Table 2: Oral glucose tolerance test of optimized formulation of *Ougeinia oojeinensis***

Groups/ Treatment	Time (min)				
	0	30	60	120	180
<b>(I) Diabetic control (Glucose)</b>	70.25 ± 4.43	168.51 ± 5.43	182.35 ± 2.86*	179.32 ± 4.85*	168.41 ± 1.56*
<b>(II) Glucose + HEOO 200mg/kg</b>	76.32 ± 4.47	188.47 ± 4.52	192.32 ± 2.74*	152.32 ± 4.72*	132.33 ± 4.34*
<b>(III) Glucose+HEOO 400mg/kg</b>	98.47 ± 3.45	192.43 ± 4.21	198.5 ± 3.54*	141.44 ± 5.85*	119.53 ± 5.23*
<b>(IV) Glucose+Phospholipids of HEOO 100mg/kg</b>	72.5 ± 3.21	171.54 ± 4.72	180.50 ± 3.71*	143.5 ± 2.52*	122.25 ± 3.83*
<b>(V) Glucose+Phospholipids of HEOO 200mg/kg</b>	90.75 ± 3.57	158.54 ± 4.74	160.7 ± 4.32*	132.5 ± 4.72*	113 ± 2.81*
<b>(VI) Glucose+ Glibenclamide</b>	78.45 ± 4.37	125.54 ± 2.85	118.72 ± 3.56*	97.35 ± 2.52*	89.35 ± 2.85*

All values represent means ± S.D of the mean (n=6) , \* p<0.05 vs diabetic control group

## **Streptozotocin-Nicotinamide Induced Diabetic Model**

### **STZ model for *Ougeinia oojeinensis***

#### **Effect of *Ougeinia oojeinensis*, phospholipids complex of *Ougeinia oojeinensis* and glibenclamide on serum glucose level in Streptozocin-nicotinamide induced diabetic rats**

The effect of 21 days treatment of HEOO (200mg/kg & 400mg/kg), phospholipid complex of HEOO (100mg/kg & 200mg/kg) and glibenclamide (10 mg/kg) on serum glucose level in streptozotocin- nicotinamide (STZ+NIC) induced diabetic rats is depicted in Table 3

The dose was introduced to the animals once a daily for 21 days. After 21 days blood samples were collected by retro-orbital plexus under mild anesthesia. Diabetic control group with no drug treatment showed no significant difference in the fasting serum glucose level after 21 days treatment as compared to the initial day treatment. However treated diabetic groups showed gradual and consistent fall in serum glucose level as shown in Figure 5.16A.

**Table 3: The effect of 21 days treatment of HEOO (200mg/kg & 400mg/kg), phospholipids of HEOO (100mg/kg & 200mg/kg) and glibenclamide (10 mg/kg) on serum glucose level in streptozotocin-nicotinamide (STZ+NIC) induced diabetic rats**

<b>Groups</b>	<b>0 day</b>	<b>7 day</b>	<b>14 day</b>	<b>21 day</b>
<b>(I) Diabetic control (Glucose)</b>	276.20 ± 5.80	274.40 ± 7.30	274.40 ± 8.75	275.00 ± 11.49
<b>(II) Glucose + HEOO 200mg/kg</b>	258.20 ± 6.62 <sup>*</sup>	247.60 ± 8.18 <sup>*</sup>	229.00 ± 7.84 <sup>*</sup>	207.40 ± 2.97 <sup>*</sup>
<b>(III) Glucose + HEOO 400mg/kg</b>	240.40 ± 4.23 <sup>**</sup>	225.08 ± 2.36 <sup>**</sup>	201.08 ± 1.95 <sup>**</sup>	199.04 ± 1.32 <sup>**</sup>
<b>(IV) Glucose + Phospholipid of HEOO 100mg/kg</b>	260.35 ± 3.42 <sup>*</sup>	221.96 ± 4.32 <sup>*</sup>	199.06 ± 2.52 <sup>*</sup>	182.56 ± 3.46 <sup>*</sup>
<b>(V) Glucose + Phospholipid of HEOO 200mg/kg</b>	246.87 ± 2.13 <sup>*</sup>	218.57 ± 3.45 <sup>*</sup>	189.34 ± 5.21 <sup>*</sup>	162.86 ± 3.40 <sup>*</sup>
<b>(VI) Glucose + Glibenclamide</b>	214.40 ± 1.82 <sup>*</sup>	197.06 ± 0.60 <sup>*</sup>	178.35 ± 2.95 <sup>*</sup>	151.70 ± 4.30 <sup>*</sup>

All values represent means ± S.D of the mean (n=6), <sup>\*</sup> p<0.05; <sup>\*\*</sup> p<0.01vs diabetic control group

### **Effect of HEOO, phospholipids of HEOO and glibenclamide on body weight in streptozotocin-nicotinamide induced diabetic rats**

The Effect of HEOO, phospholipids complex of HEOO and glibenclamide on body weight in Streptozocin nicotinamide induced diabetic rats is depicted in Table 3.

Diabetic control group with no drug treatment showed significant difference in body weight after 21 days treatment as compared to the initial day treatment. The decrease in body weight of untreated diabetic rat corresponds to fat catabolism and protein wastage and also may be due to less availability of glucose for energy utilization. However treated diabetic rat showed a significant ( $p < 0.05$ ) reduction in body weight. The diabetic rat treated with glibenclamide (10mg/kg) showed gradual and consistent increase in body weight.

### **Effect of HEOO, phospholipid complex of HEOO and glibenclamide on serum cholesterol, triglycerides, HDL-C, VLDL-C and LDL-C level in streptozotocin-nicotinamide induced diabetic rats**

After 21 days treatment, treated diabetic rat with showed significant fall in serum cholesterol, triglycerides, VLDL-C, LDL-C and increase in HDL-C level at dose of 100mg/kg and 200 mg/kg as depicted in Table 4. The standard drug glibenclamide (10mg/kg) also showed fall in serum cholesterol, triglycerides, VLDL-C, LDL-C and increase in HDL-C level as compared to control group.

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**Table 4: Effect of HEOO, phospholipid of HEOO and glibenclamide on serum cholesterol, triglycerides, HDL-C, VLDL-C and LDL-C level in streptozotocin- nicotinamide induced diabetic rats**

<b>Groups</b>	<b>Total cholesterol (mg/dl)</b>	<b>Triglycerides (mg/dl)</b>	<b>HDL-C (mg/dl)</b>	<b>LDL-C (mg/dl)</b>	<b>VLDL-C (mg/dl)</b>
<b>(I) Diabetic control (Glucose)</b>	257 ± 1.2	298 ± 2.43	42.1 ± 0.27	156.2 ± 1.12	58.7 ± 1.16
<b>(II) Glucose + HEOO 200mg/kg</b>	225.3 ± 1.41*	244 ± 1.25*	45 ± 0.13*	130.4 ± 0.52*	52 ± 2.17*
<b>(III) Glucose + HEOO 400mg/kg</b>	214 ± 1.06*	231 ± 1.76*	46 ± 1.82*	115.2 ± 0.35*	50.2 ± 1.64*
<b>(IV) Glucose + Phospholipid of HEOO 100mg/kg</b>	207 ± 2.3*	211 ± 2.84*	46.2 ± 1.04*	108.8 ± 0.26*	47.1 ± 1.32*
<b>(V) Glucose + Phospholipid of HEOO 200mg/kg</b>	191 ± 2.49*	202 ± 3.16*	47.5 ± 1.74*	102.3 ± 0.21*	44.6 ± 1.85*
<b>(VI) Glucose + Glibenclamide</b>	189 ± 1.53*	194 ± 0.31*	48 ± 2.52*	101.2 ± 0.18*	43.8 ± 1.39*

All values represent means ± S.D of the mean (n=6) , \*p<0.05 vs diabetic control group

## CONCLUSION

The present study demonstrated that *Ougeinia oojeinensis* extract could be useful in management of type-1 and type-2 diabetes associated with abnormalities in lipid profiles. The antidiabetic activity of these plants may be due to bioactive triterpenoids, steroids and flavonoids. In the nutshell, we can say that phospholipids complex based formulation could be a valuable approach to improve the therapeutic efficacy, to reduce dose and improvement in dosage regimen. Further studies need to be validated in human volunteers to claim for their antidiabetic activity.

## COMPETING INTERESTS DISCLAIMER:

Authors have declared that no competing interests exist. The products used for this research are commonly and predominantly use products in our area of research and country. There is absolutely no conflict of interest between the authors and producers of the products because we do not intend to use these products as an avenue for any litigation but for the advancement of knowledge. Also, the research was not funded by the producing company rather it was funded by personal efforts of the authors.

## References

1. Devi VK, Jain N, Valli KS. Importance of novel drug delivery systems in herbal medicines. *Pharmacogn Rev* 2010;4(7):27-31.
2. Ansari SH, Islam F, Sameem M. Influence of nanotechnology on herbal drugs: A review. *J Adv Pharm Technol Res* 2012;3(3):142-6.
3. Shaikh MS, Derle ND, Bhamber R. Permeability enhancement techniques for poorly permeable drugs: A review. *J Appl Pharm Sci* 2012;02(06):34-9.
4. Kesarwani K, Gupta R, Mukerjee A. Bioavailability enhancers of herbal origin: An overview. *Asian Pac J Trop Biomed* 2013;3(4):253-66.
5. Chaturvedi M, Kumar M, Sinhal A, Saifi A. Recent development in novel drug delivery systems of herbal drugs. *Int J Green Pharm* 2011;5(2):87-94.
6. Jain S, Jain V, Mahajan SC. Lipid based vesicular drug delivery systems. *Adv Pharm* 2014;2014:1-14.

7. Kidd PM. Bioavailability and activity of phytosome complexes from botanical polyphenols: the silymarin, curcumin, green tea, and grape seed extracts. *Altern Med Rev* 2009;14(3):226-46.
  8. Amin T, Bhat SV. A review on phytosome technology as a novel approach to improve the bioavailability of nutraceuticals. *Int J Adv Res Technol* 2012;1(3):1-5.
  9. Semalty A, Semalty M, Rawat BS, Singh D, Rawat MSM. Phytosomes: the lipid-based novel drug delivery system. *Expert Opin Drug Deliv* 2009;6(6):599-612.
  10. Fresta M, Cilurzo F, Cosco D, Paolino D. Innovative Drug Delivery Systems for the Administration of Natural Compounds. *Curr Bioact Compd* 2007;3(4):262-77.
  11. Bombardelli E, Cristoni A, Morazzoni P. Phytosomes in functional cosmetics. *Fitoterapia* 1994;65(5):387-401.
  12. Sarika D, Khar RK, Chakraborty GS, Saurabh M. Phytosomes: A Brief overview. *J Pharm Res* 2016;15(2):56-62.
  13. Afanaseva YG, Fakhretdinova ER, Spirikhin LV, Nasibullin RS. Mechanism of interaction of certain flavonoids with phosphatidylcholine of cellular membranes. *Pharm Chem J* 2007;41(7):354-6.
  14. Van Meer G, de Kroon AI. Lipid map of the mammalian cell. *J Cell Sci* 2011;124(Pt 1):5-8.
  15. Bruce A, Alexander J, Julian L, Martin R, Keith R, Peter W. *Molecular Biology of the Cell*. 4th ed. New York; Garland Sciences: 2002.
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