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# Pharmacological Investigations on polyphenol combinations for curbing diabetic and its complications

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**Abstract:**
Diabetes associated with hyperglycemia majorly affecting the vital organs due to macro and micro-vascular complications. Dietary supplement contains polyphenols possesses promising antidiabetic properties that could be useful for glycemic control and to restrain the diabetic complications. In the present study the use of epigallocatechin gallate and resveratrol alone and in combination for the treatment of diabetes for 21 days and its complications were assessed using alloxan monohyderate as a inducer (140 mg/kg). Various biochemical markers were assessed such as plasma glucose, lipid profile, kidney function test, liver glycogen, body weight and histology of kidney. Epigallocatechin and resveratrol significantly ameliorate the elevated plasma glucose and retard the dyslipidemia. Furthermore; these polyphenols significantly control the altered kidney parameters and improves the liver glycogen content and body weight. Moreover; the co-treatment of Epigallocatechin–resveratrol with glimepiride significantly modulates the glomerular sclerosis owing to its antidiabetic potential. Use of dietary supplement in the management of diabetes and its complications would be an adjuvant therapy to control hyperglycemia induced vital organ damage.

**Keywords**: Diabetes, epigallocatechin; plasma glucsoe; polyphenol; resveratrol

**Introduction**:

Globally most prominent cause of morbidity and mortality is diabetes mellitus (DM); which is a metabolic disorder characterized by increased plasma glucose which further responsible for micro and macro vascular complications affecting vital organ such as heart, kidney, nerves and eye. The main cause of DM is either insulin production defect or signaling defect i.e. insulin resistance. Earlier prevalence of DM is dramatically changes in all income group peoples/countries; there is a globally agreed target to halt the rise in diabetes and obesity by 2025. Globally 422 million peoples from low to middle income group affected with DM and

more than 1.5 million deaths were reported in each year (https://www.who.int/news-room/fact-sheets/detail/diabetes).

In past two decades researchers showed more attendtion towards natural products derived from plants such as flavonoids, glycosides, tannins, saponins etc based on their role in the treatment of various diseases and disorders.(Leonidas et al., 2017;Ong & Khoo, 2000).

Medicinal plants normally contain numerous bioactive mixtures incorporating with different targets like diabetic, cancer, hypertension etc. In ancient system of medicine Fenugreek seeds, and mulberry leaves were broadly applied to treat diabetes. Soluble dilatory fibers are possessing key role in the mamagement of diabetes like galactomannan, diosgenin, trigonelline, flavone C-glycosides ((Zentek et al., 2013).

Fruits and vegetables are overloaded with such natural polyphenols; among polyphenols flavonoids is biologically active and reported for its biological activities in inflammation, microbial infection, diabetes, hypertension, cancer and obesity. Also flavonoid possesses antioxidant potential to reduce oxidative stress in CVS and diabetes (Abotaleb et al., 2018; Graf et al., 2005).

Flavonoids are rich in food from plant origin, such as citrus fruit, grapes, apples, berries, red wine and green tea (Yuda et al., 2012). Diet containing flavonoids are beneficial in reducing the risk of multiple disease progression like diabetes, cancer, obesity and heart complications. (Nijveldt et al., 2001). Most of the flavonoids possesses some antioxidant principles therein which are responsible for these beneficial effects (Panche et al., 2016). Various flavonoids are studied for its beneficial effect in the management of various diseases and disorders like quercetin, rutin, resveratrol, gallocatachin etc.

Flavonols and anthocyanidins are both abundant in various varieties of tea and berries, respectively. Citrus fruits with high flavanone content include grapefruit, lemon, and orange. Artichokes, oregano, parsley, radish, and pepper are vegetables that are particularly high in flavones and flavonols whereas; cereals and legumes including soybean, green bean, and cowpea contains the isoflavones. Generally flavonoids are aglycon, glycoside, methylated derivatives and oligomers; best known glycosides are rutin, naringin and hesperidin (Xiao, 2017)

Catechins are natural polyphenols found in medicinal plants, and most abundant in green tea leaves (Khan & Mukhtar, 2018). Regular use of catachin rich diet would be beneficial in the management of diabetes and its complication (Khan & Mukhtar, 2018; Thielecke & Boschmann, 2009). *Camellia sinensis* is known for its antidiabetic potential based on presence of Epigallocatechin-3-gallate (EGCG) in it (Balentine et al., 1997).

Numerous studies have been conducted on the effects of tea on diabetes mellitus and its consequences. According to epidemiological studies, drinking tea raises your risk of developing type 2 diabetes and its complications (Jing et al., 2009). Furthermore, recent in vitro, in vivo, and clinical investigations have backed up tea's effects on the prevention and treatment of diabetes mellitus and its consequences (Tang et al., 2013). In addition, compared to many synthetic

hypoglycemia medications, tea is a promising hypoglycemic agent with good patient compliance fewer adverse effects and cost effective (Mahmoud et al., 2016).

Numerous studies showed the beneficial effect of Epigallocatechin in the treatment of cancer related to colon, prostate, skin, oesphagous and lung; this may be due to the induction of cancer cell apoptosis and suppression of tumor growth were discussed as a mechanism for epigallocatechin gallate (Lu et al., 2002; Ohishi et al., 2002; Yang et al., 2002)(Yang et al., 2002; Lu et al., 2002; Ohishi et al., 2002).

Furthermore, resveratrol is also studied for its antioxidant activity, antiplatelet activity, modulation of lipoprotein metabolism, suppression of tumor growth etc. by numerous researchers (Bertelli et al., 1995; Fauconneau et al., 1997; Jang et al., 1997; Soleas et al., 1997).
Multiple studies revealed the interaction of two polyphenols and cell membrane and depicted the beneficial effect of polyphenol owing to the antioxidant mechanism (Popescu, n.d.).
The aim of the present study is to check the effect of resveratrol and Epigallocatechin gallate alone and in combination on alloxan induced diabetes and its complication.

**Material and methods:**

*Experimental animals:*

Albino rats of Wistar strain weighing about 140-210 g were received from S. N. Institute of Pharmacy, Pusad, Maharashtra, India. Animals of either sex were housed under standard laboratory conditions of 22 ± 3 $^0$C temperature and relative humidity 30% and 12 h light and dark cycle with free access to standard pellet diet and water *ad-libitum.* The experimental protocol was approved by the institutional animal ethical committee SNIOP/CPCSEA/IAEC/CP-PL/20/2022.

*Acute oral toxicity study*:

The acute oral toxicity of the epigallocatachin gallate and resveratrol was performed according to the guideline set by to OECD (Organization for Economic Cooperation and development) No. 423 (OECD, 2012).

*Collection of the plant phyto-constituents:*

Epigallocatechin gallate and resveratrol were purchased from the Yucca Enterprise, Mumbai and sorted in the standard condition for further use.

*Experimental induction of diabetes in rats:*

A pre-standardized dose of alloxan monohydrate in citrate buffer was administered to the rats (140 mg/kg i.p.) and animals were kept on glucose solution (5%) to prevent possible hypoglycemia. Plasma glucose above 200 mg/dl consider as diabetic after 48 hr of alloxan administration and used of further study(Murthy et al., 2013; Navghare & Dhawale, 2016).

Antidiabetic study of co-treatment of Epigallocatechin gallate-resveratrol in alloxan-induced diabetic rats:

Diabetic animals further divided into different groups and treatment with different dosed of

Epigallocatechin gallate-resveratrol along and in combination is given along with glimepiride which is considered as standard antidiabetic agent.

Assessment of blood glucose level was performed on 1st, 7th, 14th and 21st days of the study period. The treatment design using 48 rats of either sex (42 diabetic surviving rats and 6 normal) was separated into six groups (n=6) is given below: Group I: Normal control; received suspension of 1% gum acacia (1 ml/kg, p.o.). Group II: Diabetic control; diabetic rats received suspension of 1% gum acacia (1 ml/kg, p.o.). Group III: Glim; diabetic rats received glimepiride (0.09 mg/kg, p.o.). Group IV: E 50; diabetic rats received suspension of epigallocatechin gallate in 1% gum acacia (50 mg/kg, p.o.). Group V: R 50; diabetic rats received suspension of resveratrol in 1% gum acacia (50 mg/kg, p.o.). Group VI: ER 50; diabetic rats received suspension of epigallocatechin gallate-resveratrol combination (1:1) in 1% gum acacia (50 mg/kg, p.o.). Group VII: ER 100; diabetic rats received suspension of epigallocatechin gallate-resveratrol combination combination (1:1) in 1% gum acacia (100 mg/kg, p.o.). Group VIII: ER 50 + Glim; diabetic rats received suspension of epigallocatechin gallate-resveratrol combination (1:1) in 1% gum acacia (50 mg/kg, p.o.) + glimepiride (0.09 mg/kg, p.o.).

*Evaluation of biochemical parameters:*

Assessment of the antidiabetic effect of co-treatment of resveratrol- epigallocatechin gallate was carried out by plasma glucose estimation at 0th, 7th, 14th and 21st day of treatment. Estimation of lipid profile and serum protein, albumin and creatinine were performed at the end of treatment for assessment of possible changes therein using standard diagnostic kits from Crest Diagnostic, India (Vijay et al., 2022).

7.2.6.3. *Evaluation of glycogen content in liver:*

In order to discover the effect of co-treatment of epigallocatechin gallate-resveratrol on the storage of glucose, glycogen content of liver was evaluated at the end of the experimental period. The glycogen content was measured as per the standard method described by Carroll et al., 1956 (Carroll et al., 1956).

7.2.6.4. *Change in body weight:*

Effect of co-treatment of epigallocatechin gallate-resveratrol on body weight in diabetic rats was assessed throughout the study period, i.e. at 1st, 7th, 14th and 21st day of treatment (Eleazu et al., 2013).

*b) Histological study:*

Effect of co-treatment of epigallocatechin gallate-resveratrol on histological behavior in kidney was performed on the 21st day of treatment to confirm the possible nephroprotective action of said combination. Kidneys were isolated from the sacrificed animals, cleaned with distilled water and placed in 10% formalin solution for tissues fixation and embedded in paraffin. Rotary microtome was used to cut the kidney tissues into fine sections of 5 μm thickness. Later on, these sections were treated with xylene and ethanol for deparaffinization and then stained with hematoxylin & eosin (H&E) and further examined microscopically.

7.2.7. *Statistical analysis:*

The data obtained was treated statistically by using analysis of variance (ANOVA) followed by Dunnet's test to detect any significant difference among different means, with level of significance set at $p < 0.05$. The results were expressed as Mean ± S.E.M.

Results:

Acute toxicity:

The rats that received individual epigallocatechin gallate-resveratrol combination at 5, 50 and 300 mg/kg showed no signs of toxicity during the first 30 minutes, periodically during the first 24 hours and daily thereafter, for a total of 14 days. Furthermore; epigallocatechin gallate- and resveratrol at a dose of 2000 mg/kg showed toxic event in the treated animals. All three dosed animals were observed a sign of toxicity referred to moribund or mortality in animals at each step. Based on the acute oral toxicity epigallocatechin gallate and resveratrol fall in the GSH category 4 where the $LD_{50}$ was found to be 500 mg/kg body weight. While considering acute toxicity for epigallocatechin gallate-resveratrol combination (1:1) the $LD_{50}$ was found 500 mg/kg body weight as all the three dosed animals showed toxic symptoms. In present study acute toxicity profiling gives idea about selection of doses; 10% of $LD_{50}$ was considered as the therapeutic dose for epigallocatechin gallate-resveratrol combination i.e. 50 mg/kg in 1:1proportion and two dose level were selected for further screening methods (25 and 50 mg/kg, p.o.).

**Antidiabetic study:**

Diabetic animals showed significant increase in the plasma glucose level while treatment with Epigallocatechin gallate -resveratrol alone and in combination depicted dose dependent reduction in the plasma glucose which was comparable to that of glimepiride treated animals.

Whereas; diabetic animals treated with Epigallocatechin gallate -resveratrol and glimepiride co-treatment depicted significant fall in elevated plasma glucose when compared to that of diabetic animals.

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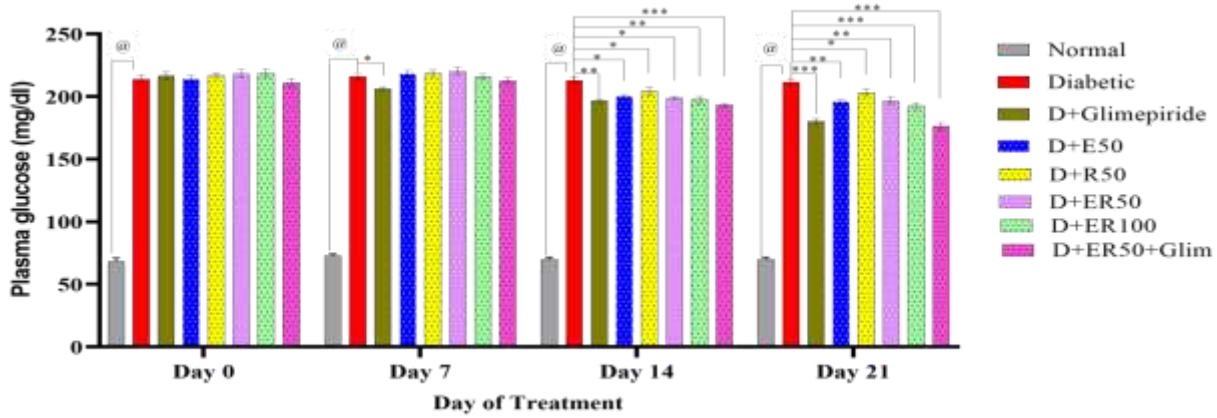


Fig1: Effect of Epigallocatechin gallate -resveratrol cotreatment on plasma glucsoe in alloxan induced diabetic animals

Values were expressed as Mean ± SEM. (n=6), ANOVA followed by Dunnett test. @p<0.001 when compared with Control; *p<0.05,**p<0.01,***p<0.001 when compared with Diabetic animals.

**Lipid profile:**

Alloxan treated diabetic animals showed significant alteration in the lipid profile in animals. Whereas; altered lipid parameters were restored by treatment with Epigallocatechin gallate -resveratrol co-treatment when compared with diabetic animals. Furthermore; diabetic animals treated with Epigallocatechin gallate -resveratrol-glimepiride co-treatment depicted significant restoration of altered lipid parameters and which were comparable to that of glimepiride treated animals.

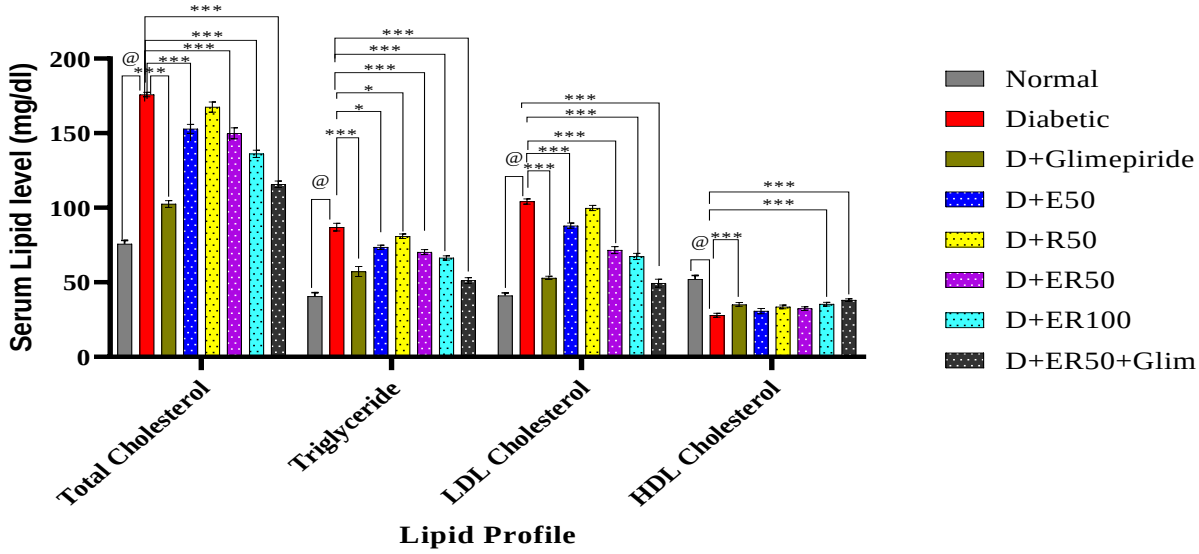


Fig2: Effect of Epigallocatechin gallate -resveratrol cotreatment on lipid profile in alloxan induced diabetic animals

Values were expressed as Mean ± SEM. (n=6), ANOVA followed by Dunnett test. @p<0.001 when compared with Control; *p<0.05,**p<0.01,***p<0.001 when compared with Diabetic animals.

| Day | Normal | Diabetic | D+Glimepiride | D+E50 | D+R50 | D+ER50 | D+ER100 | D+ER50+Glim |
|---|---|---|---|---|---|---|---|---|
| Total Protein | 8.683±0.2357 | 2.785±0.2018@ | 7.045±0.07168*** | 3.955±0.17*** | 3.177±0.1585 | 3.973333±0.16*** | 4.5±0.17*** | 4.995±0.10*** |
| Albumin | 3.810±0.1904 | 2.115±0.0502@ | 2.932±0.0975** | 2.491±0.10 | 2.157±0.2005 | 2.506±0.15 | 2.7683±0.12 | 2.866±0.12* |
| Creatinine | 0.6267±0.0583 | 2.057±0.1017@ | 1.243±0.0620*** | 1.935±0.14 | 2.138±0.0919 | 1.858±0.13 | 1.488±0.07** | 1.338±0.05*** |

**Kidney Parameters:**

Due to alloxan treatment the disturbance in the serum total protein, albumin and creatinine levels were markedly seen in the untreated diabetic animals. While animals treated with Epigallocatechin gallate -resveratrol co-treatment showed significant improvement in the altered kidney related parameters. Whereas; diabetic animals treated with Epigallocatechin gallate -resveratrol-glimepiride depicted significant changes in these altered parameters which were comparable to that of glimepiride treated animals.

Table 1: Effect of Epigallocatechin gallate -resveratrol cotreatment on kidney function test in alloxan induced diabetic animals

Values were expressed as Mean ± SEM. (n=6), ANOVA followed by Dunnett test. @p<0.001 when compared with Control; *p<0.05,**p<0.01,***p<0.001 when compared with Diabetic animals.

**Liver glycogen content:**

After induction of diabetes with the help of alloxan, the diabetic animals showed significant reduction in the level of glycogen in the liver. Diabetic animals while treating with Epigallocatechin gallate -resveratrol combination depicted improvement in the reduced glycogen content. Furthermore; the diabetic animals treated with Epigallocatechin gallate -resveratrol-glimepiride showed significant rise in the level of liver glycogen content.

Table 2 Effect of Epigallocatechin gallate -resveratrol cotreatment on liver glycogen content in alloxan induced diabetic animals

| Groups | Normal | Diabetic | D+ Glimepiride | D+E50 | D+R50 | D+ER50 | D+ER100 | D+ER50+Glim |
|---|---|---|---|---|---|---|---|---|
| Liver glycogen (mg/100g) | 13.35±0.429 | 6.236±0.632@ | 10.46±0.630*** | 8.050±0.533* | 7.088±0.272 | 8.238±0.276* | 9.213±0.459*** | 10.86±0.2201*** |

Values were expressed as Mean ± SEM. (n=6), ANOVA followed by Dunnett test. @p<0.001 when compared with Control; *p<0.05,**p<0.01,***p<0.001 when compared with Diabetic animals.

**Effect on change in body weight:**

Due to diabetes there was a significant reduction in the animal's body weight; and that reduction in the animals body weight was significantly altered by Epigallocatechin gallate-resveratrol co-treatment. Also co-treatment with Epigallocatechin gallate-resveratrol-glimepiride showed significant improvement in the reduced body weight of animals.

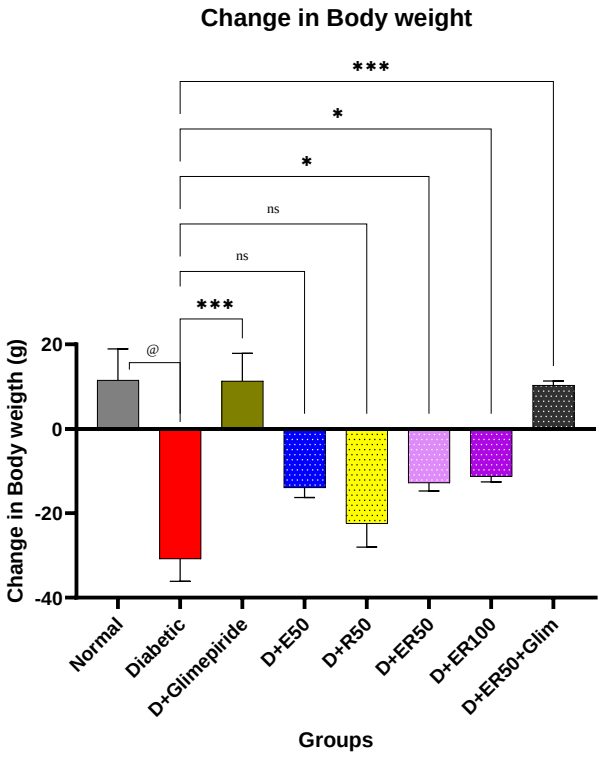


Fig 3: Effect of Epigallocatechin gallate -resveratrol cotreatment on body weight in alloxan induced diabetic animals

Values were expressed as Mean ± SEM. (n=6), ANOVA followed by Dunnett test. @p<0.001 when compared with Control; *p<0.05,**p<0.01,***p<0.001 when compared with Diabetic animals.

**Effect on Histology of Kidney**

Alloxan induced diabetes causes significant negative changes in the histolofy of kidney whereas; the diabetic animals treated with Epigallocatechin gallate-resveratrol depicted significant alteration in the archetecture of kidney while given alone and in combination with glimepiride.

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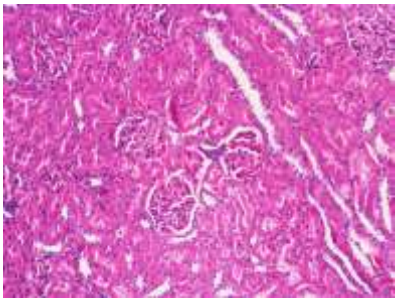
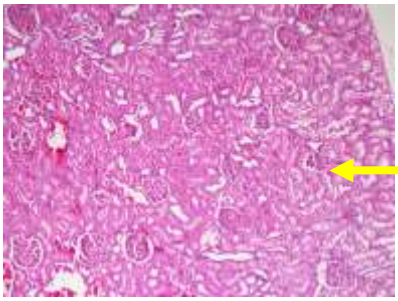
Fig 4: **Effect of Epigallocatechin gallate -resveratrol co-treatment on histology of kidney in alloxan induced diabetes in experimental animals:**

**Control**



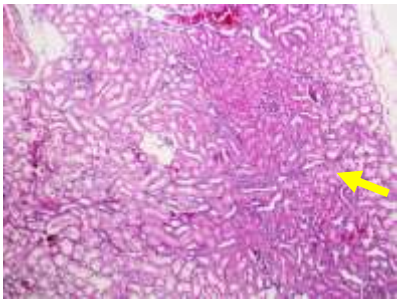
Photomicrograph of Control group kidney showing the normal archetecture of glomerulus (H & E 100X).

**Diabetic**



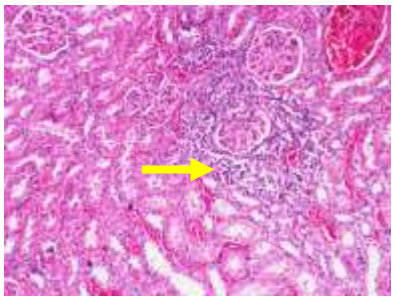
Diabetic control group kidney showed mark of Hyalinization and glomerulosclerosis (nephritis). Glomerular capillaries and tubular epithelium was affected (arrow) (H & E 100X)

**D + Glimepiride**



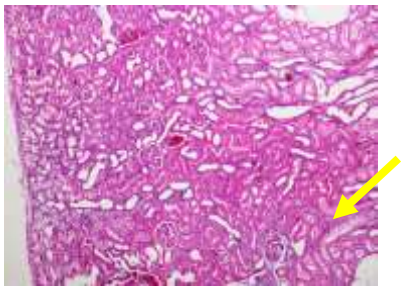
Photomicrograph of D + Glimepiride: The histological features are relatively improved compared to the non-treated diabetic group. The glomerular capillaries showed normal size (arrow). (H & E 100X)

**D+E50**



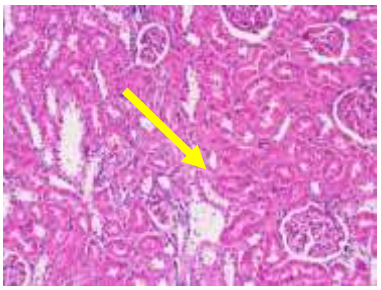
Photomicrograph of D + E50 group kidneyshowing a little mark of glomerulosclerosis (nephritis) and Hyalinization (H & E 100X) (arrow).

**D+R50**



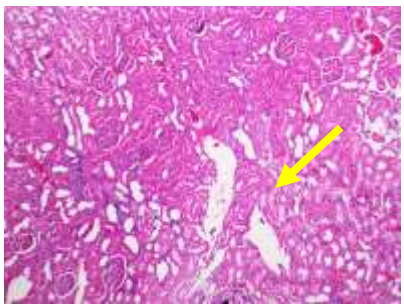
Photomicrograph of D + R50 group kidney showinga significant mark of glomerulosclerosis (nephritis) and mild hyalinization(H & E 100X)(arrow).

**D+ER50**



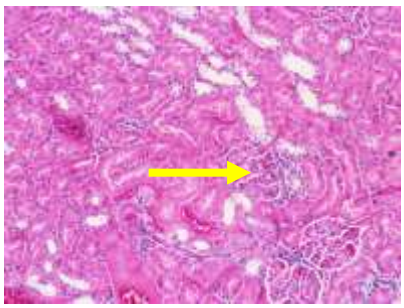
Photomicrograph of D + ER50 group kidney showing significant decline in glomerulosclerosis (nephritis) but no sign of hyalinization. (H & E 100X) (arrow).

**D+ER100**



Photomicrograph ofD + ER100 group kidney showingvery mild

**D+ER50 + Glim**



Photomicrograph of D+ ER50+ Glim group kidney showing

| | |
|---|---|
| glomerulosclerosis (nephritis) but no sign of hyalinization. (H & E 100X)(arrow). | structure of glomerulus close towards normalization. (H & E 100X) (arrow). |

**Discussion:**

Diabetes is a long-term illness that affects energy conversion and utilization from food. The majority of the food you consume is converted by your body into sugar (glucose), which is then released into your blood circulation. Pancreas releases insulin when your blood sugar levels rise. In order to utilize blood glucose for cellular metabolism and be used as energy, insulin functions like a key (*Diabetes @ Www.Cdc.Gov*, n.d.).

When you have diabetes, your body either produces insufficient insulin or uses it improperly. Too much blood sugar remains in your bloodstream when there is insufficient insulin or when cells cease reacting to insulin. That can eventually lead to major health issues like renal disease, eyesight loss, and heart disease (American Diabetes Association, 2009) .

Natural products are organic substances created by living things with biological functions, such as plants, animals, and microorganisms. Plants produce secondary metabolites with botanical origins to help them survive and grow. The production of these metabolites involves several intermediates, which are abundantly produced during Krebs cycle, glycolysis and photosynthesis (Bernardini et al., 2018; Suaifan et al., 2015).

Dietary Phenols are a complex category of bioactive secondary metabolites produced through various pathways like shikimate and acetate. Phenols are generally classified into four major groups: simple phenols or phenolic acid, coumarins, lignans, lignins, stilbenes, hydrolyzable and condensed tannins and flavonoids which include flavans, flavonols, flavones, flavanones, isoflavones, and anthocyanins (Kumar & Goel, 2019; Sun et al., 2020). Several simple phenolic acids were assessed clinically and in vivo for their antidiabetic activities.

Dietary phenols possesses certain properties which protect the pancreatic beta cells and reduce cellular apoptosis along with improvement in beta cell proliferation and reduces reactive oxygen species induced damage, furthermore; dietary phenol activates insulin secretion, insulin signaling, alteration in inflammation response, and subsequent inhibition of AGE formation (Sun et al., 2020).

Green tea was known for its antidiabetic potential from past few decades. EGCG found in green tea was reported for its antidiabetic potential by protecting and restoring the pancreatic beta cells from alloxan induced damage and reducing the plasma glucose concentration (Bone et al., 1985). Moreover; EGCG was medicated antidiabetic activity by promoting the insulin secretion and also possesses insulinomimetic activity (Ortsäter et al., 2012). Epigallocatechin gallate is known as an antioxidant, but recent investigations have revealed other direct actions independent from it, such as anti-hypertrophy, anti-inflammatory, anti-myocardial infarction and anti-atherosclerosis (Khan & Mukhtar, 2018; Kim et al., 2014)

Certain plant and fruits such as grapes, peanuts, mulberries, and blueberries are rich source of natural antioxidant component Resveratrol (Burns et al., 2002; Rimando et al., 2004).

Resveratrol is known to induce, among others, anti-oxidative (Frombaum et al., 2012), cardio-protective (Zordoky et al., 2015), anti-cancer (Carter et al., 2014; Singh et al., 2015), anti-inflammatory (Poulsen et al., 2015), neuro-protective (Bastianetto et al., 2015; Pallàs et al., 2009) and anti-obesity (de Ligt et al., 2015) effects. Resveratrol's anti-diabetic activity has been thoroughly investigated in past few years using various animal models, pointing to its lucrative effects towards diabetes and its complications.

Alloxan monohydrate causes increase in plasma glucose within three hours of administration in experimental animals. Alloxan damages the pancreatic beta cells by producing reactive oxygen species which attacks on beta cells and inhibit its function to secrete insulin (Lenzen, 2008; Szkudelski, 2001). A structural similarity to that of glucose transport of alloxan via GLUT2 channels is facilitated in beta cells. Since both glucose and alloxan due to similarity in molecular shape competes for the same transport protein, it therefore implies that high level of glucose in circulation will automatically lower the chance of alloxan binding to GLUT2. Even at a relatively equal concentration of both molecules, GLUT2 has a stronger affinity for glucose than alloxan and thus favors the binding of glucose compared with alloxan (Elsner et al., 2002).

Diabetic animals showed significant rise in the plasma glucose that can be confirmed by comparing the plasma glucose with that of non diabetic animals, this rise in the plasma glucose was may be due to destruction of pancreatic beta cells further leads to reduced production of insulin that may lead to hyperglycemia by alloxan monohydrate (Szkudelski, 2001).

Diabetic animals treated with Epigallocatechin gallate and resveratrol significantly attenuate the rise in plasma glucose which was comparable to that of glimepiride treated animals (Fig 1).

Numerous information available suggested that Epigallocatechin gallate possess antidiabetic property and it significantly lowers plasma glucose (Kao et al., 2000). In present study Epigallocatechin gallate significantly lowers plasma glucose that may be due to its insulinomimetic action and reduces glucose production in H4IIE cells and reduction in the genes expression in PEPCK and G6Pase genes that regulate gluconeogenesis (Waltner-Law et al., 2002). Alloxanized animals treated with resveratrol significantly lower the plasma glucose when compared to diabetic animals that may be its antioxidant property to protect the pancreatic beta cell from oxidative damage by reducing ROS production via chelation at the binding site and increasing endogenous antioxidant enzyme level (Gülçin, 2010; Leonard et al., 2003). Furthermore; diabetic animal treated with combination Epigallocatechin gallate and resveratrol possesses promising effect on elevated plasma glucose due to their promising antioxidant and protective effect on pancreatic beta cells.

Epigallocatechin gallate and/or in combination with resveratrol displayed more promising antidiabetic impact as compared to that of Epigallocatechin gallate and resveratrol, which indicates that Epigallocatechin gallate and resveratrol has high anti-oxidant characteristics.

Uncontrolled diabetes may responsible for alteration in the serum total cholesterol, triglyceride, LDL cholesterol and HDL cholesterol levels. Alloxan induced diabetes depicted disturbance in the normal lipid profile of animals when compared to that of non-diabetic animals.

Whereas; diabetic animals treated with Epigallocatechin gallate and resveratrol showed promising alteration in the disturbed lipid profile by increasing the HDL cholesterol level and decrease in serum total cholesterol, triglycerides and LDL cholesterol levels.

Whereas; treatment with Epigallocatechin gallate and resveratrol alone and in co-treatment depicted significantly reorganize the diabetic state by reducing the levels of serum total cholesterol, triglyceride and LDL cholesterol and enhancement in serum HDLc levels (Fig 2). Epigallocatechin gallate and resveratrol known for reduction in the levels of serum lipid in adiposities in the presence or absence of insulin was already reported (Szkudelska et al., 2009). EGCG alone has the potential ability to decrease the serum lipid biomarkers by increasing the fat metabolism through oxidation increase fat oxidation (Hodgson et al., 2013).

Diabetes may responsible for chronic kidney disease as there is rise in plasma glucose that further responsible for glomerular sclerosis; and glycemic control prevents the chances of kidney lesions (Fig 4). In hyperglycemic state there is formation of AGEs which mainly responsible for cellular injury and further alter the gene function which may cause the increment in the cellular matrix (Shahbazian & Rezaii, 2013). Diabetic animals treated with Epigallocatechin gallate and resveratrol ameliorate the altered kidney parameters significantly when compared to that of diabetic animals (Table 1).

Additionally to the hyperglycemia and dyslipidemia the diabetic animals showed significant fall in the liver glycogen content when compared to that of normoglycemic animals. Treatment with Epigallocatechin gallate and resveratrol alone and in co-treatment showed significant alteration in liver glycogen content, furthermore; the animals treated with Epigallocatechin gallate, resveratrol and glimepiride co-treatment depicted marked rise in liver glycogen content when compared to that of diabetic animals (Table 2). Epigallocatechin gallate was already reported for it beneficial effect in diabetes by improving the insulin resistance and promoting the glycogen synthase kinase activity which mainly responsible for improvement in the liver glycogen content (Chen et al., 2018). Furthermore, resveratrol also reported for its antidiabetic effect and also depicted rise in liver glycogen content via improving the glycogen synthase and decrease in glycogen phosphorylase activity (Pallàs et al., 2009).

Diabetes always associated with reduction in the body weight as alloxan treatment showed significant reduction in the body weight when compared to non-diabetic animals. Diabetic animals treated with Epigallocatechin gallate and resveratrol depicted significant improvement in the reduced body weight in diabetic animals (Fig 3). Treatment with Epigallocatechin gallate and resveratrol significantly ameliorate the reduction in body weight most probably by improving the glycemic control (Rašković et al., 2019; Zhu et al., 2022). Furthermore diabetic animals treated with Epigallocatechin gallate, resveratrol and glimepiride co-treatment significantly improves the body weight.

**Conclusion**:

In present study the effect of Epigallocatechin gallate and Resveratrol were assessed using alloxan induced diabetes and possesses remarkable antidiabetic potential and which were

comparable to that of glimepiride treated animals. The effects of Epigallocatechin gallate and Resveratrol were confirmed based on the alteration in the dyslipidemia and amelioration of altered kidney parameters in diabetes when compared to disease control animals. Furthermore; Epigallocatechin gallate and Resveratrol improves the liver glycogen content and improvement in the body weight of diabetic animals that may be due to improvement in the glycemic control. While considering the effect of co-treatment of Epigallocatechin gallate -Resveratrol and glimepiride depicted significant antidiabetic activity and also reduces the various complications associated with diabetes. Further; mechanistic studies are required to find out the exact mechanism behind the glucose lowering, controlling dyslipidemia and altered kidney related parameters of Epigallocatechin gallate and Resveratrol.

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