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Anti-hyperglycemic activity of methanolic extract of *Flacourtia montana* in streptozotocin induced diabetic rats

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Abstract
Diabetes mellitus is a chronic metabolic disorder characterized by hyperglycemia resulting from defects in insulin secretion, insulin action, or both. More than 200 species of plants possess antidiabetic properties which were evaluated mostly by screening tests and some are yet to explore. One such plant named, *Flacourtia montana* (Family-Flacourtiaceae) was used in these study to evaluate anti-hyperglycemic activity of methanolic leaf extract in STZ induced type I diabetes model. Animals were randomized in to five groups of six each, all the groups except Group I were made diabetic by intraperitoneal injection of Streptozotocin (STZ) at the dose rate of 45 mg/kg body weight. The Group I served as the normal control, Group II and III as STZ and Glibenclamide control. The group – IV and group V were administered methanolic leaf extract @ 100 mg/kg and 250 mg/kg body wt for 15 days. The result revealed an increase in Blood glucose, Triglycerides, Total Cholesterol, LDL-C, AST, ALT level and decrease in HDL-C and non-significant decrease in body weight in all groups except normal control on day zero. The group IV and V exhibited reduced blood glucose level and all other parameters in 15 days of treatment. Thus, the result revealed the anti-hyperglycemic activity of methanolic leaf extract of *F. montana*. Further analytical experiment is needed to confirm exact mechanism of action.

Keywords: Diabetes mellitus, *Flacourtia montana*, anti-hyperglycemic activity and blood glucose, triglycerides, total cholesterol and lipid profile

1. Introduction
Diabetes mellitus (DM) is a chronic metabolic disorder characterized by a high blood glucose concentration (hyperglycaemia) caused by [fasting plasma glucose ≥ 7.0 mmol/l (126mg/dl) or 2–h post prandial plasma glucose ≥ 11.1mmol/l (200mg/dl) following a 75g oral glucose load or random plasma glucose ≥ 11.1mmol/l (200mg/dl) in the presence of diabetes symptoms] insulin deficiency, often combined with insulin resistance (WHO, 2006). The consequences of diabetes include microangiopathy like nephropathy, neuropathy, retinopathy and macrovascular diseases like accelerated atheroma, stroke, myocardial infarction and cardiovascular disorders. It was known to ancient Indian physicians as 'Madumeha'. (Fakeye *et al.*, 2007). Diabetes imposes an increasing economic burden on national health care systems worldwide and an estimated global health expenditure on diabetes is 490$ billion by 2030 (Fonseca, 2006) [2]. From the public health standpoint, the only cost-effective way of dealing with diabetes is to prevent it.
Treatment of DM consists of diet management and/or insulin therapy. The therapy attempt to normalize metabolic activities, especially blood glucose levels. The pharmacological treatment of DM is based on two types of drug *viz*, Insulin and Oral Hypoglycemic agents. The insulin sensitizers like Thiazolidinediones /glitazones, insulin secretogogues like sulfonylurea and glinides, glucose metabolism enhancers alpha-glucosidase inhibitors, incretin mimetics and glucose dependent Insulinotropic polypeptide, incretin enhancers and new category like DPP-4 inhibitors are the major oral hypoglycemic agents in use (Cramer *et al*., 2008, Lorenzati *et al*, 2010, Sandoval and Sisley, 2015) [3, 4, 5]. Even though most of the oral antihyperglycemic agents decrease the risk of complication in diabetes. They are expensive and have many adverse effects including hypoglycemia and obesity (Sharma *et al.*, 2008) [6, 15]. The management of diabetes is a challenge to modern medicine as allopathic medicine has limited application in alleviation of pancreatic ailments.
However, traditional herbal medicines have a long history of use in the treatment of pancreatic diseases, though most of these agents lack scientific validation.
Herbal remedies are potent source of drugs for the treatment of diabetes mellitus in Indian system of medicine and other ancient systems of the world.

Herbal remedies are potent source of drugs for the treatment of diabetes mellitus in Indian system of medicine and other ancient systems of the world. India is a country with a vast reserve of natural sources and rich history of traditional medicine. There are more than 1200 varieties of living species, identified with anti-diabetic property. Traditional medicines from available medicinal plants offer great potential for the discovery of new antidiabetic drugs (Jung *et al.,* 2006). Research on phytochemicals for therapy of DM is an important facet of biomedical research all over the world. Anti-hyperglycemic activity of these herbal formulations is due to the ability to restore the function of pancreas by either an increase in insulin output or inhibition of the intestinal absorption of glucose. Herbal anti-diabetic therapy is economical, easily available and with limited side effects during prolonged administration. All these highlights the importance for an alternative therapy, with plant derived drugs. An ideal oral treatment for diabetes is a drug that controls the glycemic level and prevents development of microangiopathy and macrovascular complications in cardiovascular and renal systems associated with diabetes mellitus.

Plants of *Flacurtia Spp*. are endemic to Kerala and grows in semi-evergreen and moist deciduous forests of Western Ghats, up to 1000 m (1800 m). There are reports on fruits of *Flacourtia indica*, being used as appetizer, digestive tonic, diuretic, in jaundice and enlarged spleen. Barks are used for the treatment of intermittent fever and roots are used in nephritic colic and gum in cholera (Joy *et al*., 2001). Anti-hyperglycemic effect of *Flacourtia jangomas* was demonstrated in STZ induced diabetes rats by (Singh *et al.,* 2010). Other pharmacological investigation includes the evaluation of antihistaminic activity of ethanolic extract of leaves of *Flacurtia indica* in experimental guinea pig model (Tyagi *et al*., 2011). The antimicrobial activity of *Flacurtia montana* (Sarker *et al*., 2011) and hepatoprotective/'/ activity of *Flacurtia indica* (Nazneen *et al.,* 2009) were also reported but literatures are scarce on anti-hyperglycemic activity of *F. montana*.

Hence the preset study aims to evaluate the anti-hyperglycemic activity of methanolic leaf extract of *F. montana* in STZ induced diabetic rats.

2. Materials and Methods

2.1 Collection and identification of plant materials

Fresh leaves of *F. montana* were collected from the district of Wayanad. From the collected leaves, herbarium was prepared for identification and authentication in the Department of Botany, University of Calicut and voucher specimen was deposited with accession number 3197.

2.2 Preparation of test materials

Fresh leaves of *Flacurtia montana* were collected and shade dried. Dried leaves were coarsely powdered to a mesh size of 40-50 in an electrically driven pulverizer and sieved to remove the coarse substances. They were made into thimbles and weighed. Extraction was done using methanol in a Soxhlet extraction assembly until the solvent turned clear in the Soxhlet. The liquid extract obtained was collected in round bottom flask and solvent was evaporated by using Rotary Vacuum Evaporator (M/s Buchi, Switzerland) under reduced pressure. The semisolid extract obtained was air dried and stored in the refrigerator at 4˚C for further use.

2.3 Phytochemical analysis

Methanolic extract of leaf of *F. montana* were analyzed qualitatively for various phytochemical constituents as per standard procedures mentioned by Harborne (2005).

2.4 Evaluation of acute oral toxicity

Acute oral toxicity testing of methanolic extract of leaf and fruit juice was carried out according to the Organization for Economic Co-operation Development [OECD, TG-420 (2001)] guidelines.

2.5 Experimental animals

The antidiabetic study was conducted in 30 adult albino Wister rats of either sex weighing 200 – 230 g. The experiment was approved by the Institutional Animal Ethics Committee (IAEC). All the animals were maintained in well ventilated cages in the laboratory under standard management conditions for one week, to get acclimatized with new laboratory environment, before the commencement of the experimental setup. The experiment was carried out for a period of 20 days.

2.6 Protocol for induction of diabetes

All the experimental animals were fasted overnight. Their body weight was noted and blood glucose was estimated on the next day morning (0th day). Diabetes was induced in all the treatment group except the normal control by intraperitoneal administration of freshly prepared Streptozotocin (STZ) at the dose rate of 45 mg/kg body weight (Srinivasan and Ramarao, 2007; King, 2012). On 5th day, blood glucose was estimated by method of GOD-POD glucose kit. Rats showing moderate hyperglycemia (200-350 mg/dl) were selected for study and grouped randomly.

A weighed quantity of the methanolic extract was homogenized with distilled water and was administered orally using oro-gastric catheter to individual rats for 15 days at the dose rate of 100 mg/kg body weight or 250 mg/kg body weight. The reference drug for DM evaluation in type-1 diabetic rat model, second generation Sulfonyl urea, Glibenclamide was used as a positive control in the present study. Tablet Daonil® (Glibenclamide) 5 mg was powered and dissolved in distilled water so that 5 ml contained 5 mg of Glibenclamide. It was given orally at the dose rate of 5 mg/kg from 5th day to 15th day as described.

Table 1: experimental design to conduct anti-hyperglycemic activity of *F. montana*.

Group		Treatment
Group I	Normal control	Animals receiving vehicle distilled water orally for 15 days.
Group II	STZ control	STZ treated animals receiving the vehicle orally for 15 days
Group III	Glibenclamide control	STZ treated animals receiving Glibenclamide at 5 mg/kg body weight orally for 15 days.
Group IV	Test 1	STZ treated animals receiving leaf extract of *F. montana* at 100 mg/kg B.W. orally for 15 days.
Group V	Test 2	STZ treated animals receiving leaf extract of *F. montana* at 250 mg/kg B.W. orally for 15 days.

From day 5 of STZ administration, animals were treated orally daily with the test substance. Blood samples were collected on day zero, day 7 and day 15 for estimating blood glucose. Also serum was separated for estimation of TG, TC, HDL-C, LDL-C and serum marker enzymes like AST and ALT. Body weight was also recorded on these days. The animals were sacrificed on day 16, pancreas and liver were collected. Pancreas was preserved in 10 per cent neutral buffered formalin for histopathological examination. Liver tissue was used for the estimation of glycogen content.

3. Results

The results of phytochemical analysis are summarized in table 2.

Table 2: Qualitative phytochemical screening of methanolic leaf extract of *F. montana.*

S. No	Active phytoconstituents	Methanolic leaf extract of *F. montana*
1	Alkaloids	Present
2	Flavonoids	Present
3	Glycosides	Present
4	Steroids	Absent
5	Tannins	Present
6	Phenolic compounds	Present
7	Terpenes	Present
8	Carbohydrates	Present
9	Saponins	Present
10	Gums and mucilages	Absent

An acute oral toxicity study of extract was performed in rats after a single dose administration as per OECD guidelines [OECD, TG-420 (2001)]. The results of the acute oral toxicity confirm that the leaf extract possessed no toxicity at dose 2000 mg/kg. The animals were healthy, feed and water intake was normal. There was no behavioural changes during the entire period of study.

3.1 Evaluation of general health improvement and anti-hyperglycemic effect of *F. montana*

The body weight was recorded before administration of STZ, after induction of diabetes (zero day), during the course of treatment (7th day) and at the end of the treatment (14th day). The gain in body weight was taken as a criterion to assess the health status. The liver glycogen level which indicated the energy reserve in body was also estimated on 15th day as a part of assessment of the health status of treatment groups. The blood glucose level was estimated before and after the induction of diabetes, during the course of the treatment (7th day) and at the end of the treatment (14th day) to assess the anti-hyperglycemic effect of methanolic leaf extract *F. Montana*

3.2 Body weight

Table 3: Effect of the methanolic leaf extract of *F. montana* on the body weight of rats, g.

Group	Day - 0	Day - 7	Day – 14
I	$103.35\pm0.7^{ns,B}$	$103\pm0.77^{ns,D}$	$103.17\pm0.65^{ns,E}$
II	$256.45\pm8.42^{ns,A}$	$258\pm7.63^{ns,A}$	$258\pm7.43^{ns,A}$
III	$267.22\pm6.42^{a,A}$	$136.33\pm0.61^{b,C}$	$123.33\pm0.49^{c,D}$
IV	$261.09\pm7.55^{a,A}$	$162.83\pm0.83^{b,B}$	$139\pm0.58^{c,C}$
V	$243.76\pm7.04^{a,A}$	$143.83\pm0.7^{b,C}$	$128\pm0.63^{c,D}$

n=6, P< 0.05, expressed as mean ± S.E. Means with atleast one common superscript (a-c) for rows and (A-C) for columns do not differ significantly at 5 % level.

3.3 Blood glucose level

The mean values indicating the reduction of blood glucose are represented in table 4

Table 4: Effect of the methanolic leaf extract of *F. montana* on the blood glucose level, mg/dl

n=6, P< 0.05, expressed as mean ± S.E. Means with atleast one common superscript (a-c) for rows and (A-D) for columns do not differ significantly at 5 % level.

The results indicated that the methanolic leaf extract at dose rate of 250 mg/kg is as potent as glibenclamide at dose of 5 mg/kg. In lowering elevated blood glucose level even though the extract at lower doses reduced the elevated blood glucose level, the effect was not as pronounced as with the higher dose of methanolic extract.

3.4 Total serum cholesterol level

The effect of various treatments on the total serum cholesterol level for groups I to VII on zero day, 7th day and 14th day are presented in table 5

Table 5: Effect of the methanolic extract of *F. montana* on total cholesterol, mg/dl

Group	Day - 0	Day - 7	Day - 14
I	$87.96\pm1.08^{ns,B}$	$87.67\pm1.02^{ns,E}$	$88\pm1.13^{ns,F}$
II	$141.09\pm0.75^{ns,A}$	$141\pm0.82^{ns,A}$	$140.83\pm0.79^{ns,A}$
III	$139.6\pm0.67^{a,A}$	$112.83\pm0.79^{b,D}$	$93.33\pm0.56^{c,E}$
IV	$140.87\pm0.58^{a,A}$	$133.67\pm0.56^{b,B}$	$123.67\pm0.49^{c,C}$
V	$141.97\pm0.6^{a,A}$	$124.33\pm1.69^{b,C}$	$108.17\pm0.95^{c,D}$
Group	**Day - 0**	**Day - 7**	**Day - 14**
I	$218.00 \pm 0.72^{ns,A,B}$	$217.67 \pm 0.84^{ns,A}$	$217.67 \pm 0.84^{ns,A}$
II	$224.633 \pm 1.91^{a,A}$	$211.67 \pm 0.71^{b,B}$	$205.00 \pm 0.96^{c,B,C}$
III	$211.550 \pm 1.60^{a,c,B,C}$	$208.50 \pm 1.43^{b,c,B,C}$	$208.83 \pm 1.32^{b,B}$
IV	$211.700 \pm 1.32^{a,c,B,C}$	$206.33 \pm 0.76^{b,C}$	$201.50 \pm 0.95^{c,C}$
V	$209.950 \pm 1.41^{a,c,C}$	$205.83 \pm 0.91^{b,C}$	$201.83 \pm 0.65^{c,C}$

n=6, P< 0.05, expressed as mean ± S.E. Means with atleast one common superscript (a-c) for rows and (A-F) for columns do not differ significantly at 5 % level.

3.5 Serum triglycerides

The effect of various treatment in serum triglyceride level on day zero, 7 and 14 results of serum triglycerides level analysis is presented in table 6.a

Table 6: Effect of the methanolic extract of *F. montana* on serum triglycerides, mg/dl

Group	Day - 0	Day - 7	Day - 14
I	$107.85\pm0.74^{ns,D}$	$107.5\pm0.76^{ns,F}$	$107.67\pm0.67^{ns,E}$
II	$168.25\pm1.87^{ns,A,B}$	$167.67\pm1.86^{ns,A}$	$166.5\pm2.26^{ns,A}$
III	$174.71\pm2.05^{a,A}$	$125.67\pm0.56^{b,E}$	$114.67\pm1.63^{c,D}$
IV	$160.3\pm0.95^{a,A,C}$	$146.17\pm1.17^{b,C}$	$129.33\pm0.61^{c,C}$
V	$163.42\pm1.22^{a,B,C}$	$135.83\pm0.79^{b,D}$	$125.17\pm0.4^{c,C}$

n=6, P< 0.05, expressed as mean ± S.E. Means with atleast one common superscript (a-c) for rows and (A-D) for columns do not differ significantly at 5 % level.

3.6 High Density Lipoprotein-Cholesterol (HDL-C) level

The effect of various treatments on serum HDL-C level is summarized on table 7.

Table 7: Effect of the methanolic extract of *F. montana* on the HDL-C, mg/dl

Group	Day - 0	Day - 7	Day - 14
I	$62.96\pm0.63^{ns,A}$	$62.17\pm0.6^{ns,A}$	$62.17\pm0.6^{ns,A}$
II	$51.36\pm0.59^{a,A,B}$	$42.67\pm0.67^{b,D}$	$33.17\pm0.6^{c,E}$
III	$53.04\pm0.8^{b,A}$	$51.83\pm1.11^{b,C}$	$59.67\pm0.42^{a,B}$
IV	$50.94\pm0.6^{b,A,B}$	$55\pm0.26^{a,B}$	$56.5\pm0.22^{a,C,D}$
V	$50.63\pm0.7^{c,A,B}$	$55.83\pm0.48^{b,B}$	$57.67\pm0.33^{a,C,B}$

n=6, P< 0.05, expressed as mean ± S.E. Means with atleast one common superscript (a-c) for rows and (A-E) for columns do not differ significantly at 5 % level.

3.7 Low Density Lipoprotein-Cholesterol (LDL-C) level

The effect of various treatments on serum LDL-C level is summarized on table 8

Table 8: Effect of the methanolic extract of *F. montana* on the LDL-C, mg/dl

Group	Day - 0	Day - 7	Day - 14
I	$32.10\pm0.49^{ns,C}$	$32.03\pm0.58^{ns,E}$	$31.5\pm0.67^{ns,F}$
II	$43.54\pm0.54^{ns,A,B}$	$43.17\pm0.31^{ns,A}$	$43.75\pm0.37^{ns,A}$
III	$43.36\pm0.34^{a,A,B}$	$37.01\pm0.77^{b,D}$	$33.33\pm0.71^{c,E}$
IV	$41.31\pm0.54^{a,B}$	$37.67\pm0.21^{b,C,D}$	$36.12\pm0.26^{c,C,D}$
V	$41.64\pm0.54^{a,B}$	$36.17\pm0.48^{b,D}$	$34\pm0.68^{c,D,E}$

n=6, P< 0.05, expressed as mean ± S.E. Means with atleast one common superscript (a-c) for rows and (A-F) for columns do not differ significantly at 5 % level.

3.8 Serum Aspartate aminotransferase (AST)

The serum aspartate aminotransferase of groups I to VII on zero day, 7th and 14th day are presented in table 9.

Table 9: Effect of the methanolic extract and fruit juice of *Flacortia montana* on AST level, IU/L

Group	Day - 0	Day - 7	Day - 14
I	$65.22\pm0.56^{a,B}$	$54.67\pm0.49^{b,E}$	$64.83\pm0.54^{a,F}$
II	$87.71\pm0.46^{ns,A}$	$87.83\pm0.48^{ns,A}$	$87.83\pm0.48^{ns,A}$
III	$87.01\pm0.38^{a,A}$	$70.67\pm0.42^{b,D}$	$60.17\pm0.48^{c,E}$
IV	$88.1\pm0.42^{a,A}$	$83.33\pm0.56^{b,B}$	$79.17\pm0.48^{c,C,D}$
V	$87.47\pm0.63^{a,A}$	$80.5\pm0.43^{b,C}$	$75.33\pm0.33^{c,E,D}$

n=6, P< 0.05, expressed as mean ± S.E. Means with atleast one common superscript (a-c) for rows and (A-F) for columns do not differ significantly at 5 % level.

3.9 Serum Alanine aminotransferase (ALT)

The serum alanine aminotransferase values for groups I to VII on zero day, 7th and 14th day are presented in table 10.

Table 10: Effect of the methanolic extract and fruit juice of *F. montana* on ALT level, IU/L

Group	Day - 0	Day - 7	Day - 14
I	$20.52\pm0.27^{ns,D}$	$20.33\pm0.33^{ns,F}$	$20.67\pm0.33^{ns,F}$
II	$43.34\pm0.22^{ns,A}$	$43.5\pm0.22^{ns,A}$	$43.5\pm0.22^{ns,A}$
III	$42.57\pm0.32^{a,A,B}$	$31.17\pm0.54^{b,D,E}$	$25\pm0.37^{c,E}$
IV	$41.36\pm0.63^{a,B,C}$	$35.17\pm0.54^{b,C}$	$29.5\pm0.43^{c,C}$
V	$40.98\pm0.43^{a,C}$	$33.83\pm0.4^{b,D}$	$28.17\pm0.31^{c,D}$
VI	$42.96\pm0.61^{a,A,B}$	$37.67\pm0.56^{b,B}$	$35\pm0.26^{c,B}$
VII	$42.68\pm0.37^{a,A,B}$	$36.17\pm0.31^{b,B,C}$	$32.17\pm0.31^{c,C}$

n=6, P< 0.05, expressed as mean ± S.E. Means with atleast one common superscript (a-c) for rows and (A-F) for columns do not differ significantly at 5 % level.

3.10 Liver glycogen level

Liver glycogen was estimated, after sacrificing the animals on last day of the treatment and is presented in table 11.

Table 11: Effect of the methanolic extract and fruit juice of *Flacortia montana* on the liver glycogen, gram%

Group	Liver Glycogen
I	4.891667 ± 0.71^{nsA}
II	2.401667 ± 0.68^{aF}
III	3.993333 ± 0.73^{aB}
IV	3.708333 ± 0.59^{aC}
V	3.961667 ± 0.64^{aB}

n=6, P< 0.05, expressed as mean ± S.E. Means with atleast one common superscript (a-c) for rows and (A-F) for columns do not differ significantly at 5 % level

3.11 Histopathological examination of pancreas

The results of histopathology are represented in plate II and III.

The pancreas showed normal histoarchitecture with well-developed globules of acini, normal islet cells, without fibrosis or inflammation in group I. In diabetic rats the islet cells showed atrophy with inflammatory edema, necrosis and fibrotic changes. There was remarkable regeneration of pancreatic cells with normal islet cells, prominent nucleus and nucleolus in the treatment groups in a dose dependent manner. The pancreas of glibenclamide treated rats showed minimal necrosis and mild atrophy with minimum fibrotic changes.

4. Discussion

Diabetes mellitus is an endocrine disorder involving derangements in the metabolism of carbohydrates, fats and proteins. This disease is a pandemic in both developed and developing countries. The present treatment regimen of diabetes include exercise, diet modification and use of oral hypoglycemic agents and insulin therapy. Although many drugs are available for the management of diabetes, they are usually expensive and have adverse effects like hypoglycemia and obesity (Sharma *et al.*, 2008) [6, 15]. Screening of herbs for anti-hyperglycemic activity is of great significance in this context.

Diabetes is associated with weight loss as a consequence to elevated muscle wasting and decrement of tissue proteins (Chatterjee and Shinde, 2002). In our study, there is marked reduction in body weight in the DM group, whereas the decrease in treated group was dose dependent.

There was a dose dependent reduction in the serum blood glucose values on day 7 and 14 on all the treatment groups. Sulphonylurea such as glibenclamide stimulate insulin secretion from pancreatic β cells principally by inhibiting ATP-sensitive K+ channels (Watkins 2003). The effect of

glibenclamide and methanolic extract of leaf of *F. montana* at 250 mg/kg on day 14 were comparable at $p<0.05$. This indicates that the methanolic extract contains potent phytochemicals that can reduce the elevated blood glucose.

The post prandial hyperglycemia is usually controlled by liver by increasing the synthesis of glycogen and decreasing glycogenolysis. The marked reduction in liver glycogen level was observed during 15 days study in STZ induced diabetic animals in the group II whereas, group III regained the glycogen level to normal. The group IV and V which are treated with methanol leaf extract of *F. montana* remarkably increased the glycogen level in liver. Test substance can reduce blood glucose level by several potential mechanisms. It is well established that, the blood glucose lowering activity can be mediated via converting glucose to glycogen by glycogen synthetase and promotion of glycogen storage in liver and in skeletal muscles (Mera, 1997; Jayakody and Ratnasooriya, 2008). Which is evident in present study by significant increase in liver glycogen level in extract treated groups.

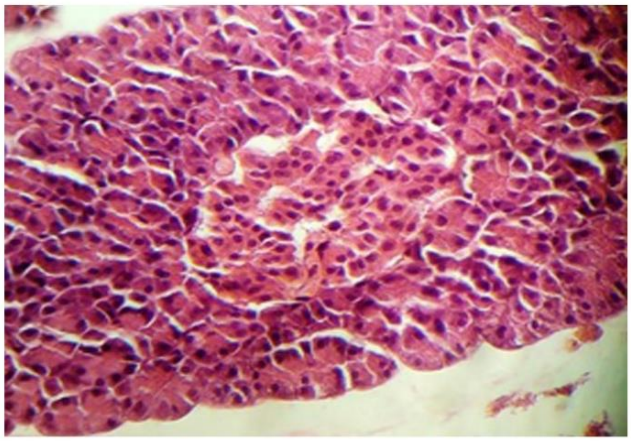
Qualitative phytochemical screening of methanolic leaf extract and fruit juice of *F. montana* showed the presence of alkaloids, glycosides flavonoids, tannins, terpenes, phenolic compounds, carbohydrates and saponins. These constituents may in part be responsible for the observed significant activity of this extract and fruit juice, either single or in synergy.

The bioactive plant metabolites are noted for potential as antioxidants and free radical scavengers (Kusirisin *et al*., 2009). There are several reports suggesting that phytochemicals like terpenes, saponins, tannins and alkaloids are responsible for the antidiabetic activity. (Kumar *et al.*, 2011). Most of the beneficial health effects of flavonoids are attributed to their antioxidant and chelating abilities. By virtue of their capacity to inhibit LDL oxidation, flavonoids have demonstrated unique cardioprotective effects (Mazur et al., 1999).

Some flavonoids such as rutin, myricetin, kaempferol and quercetin have been previously reported to inhibit α-glucosidase which has a prominent role in carbohydrate digestion. These flavonoids exhibit both hypoglycemic and antioxidant effects in diabetic animals (Kamalakkannan and Prince, 2006; Wang *et al.*, 2013).

Some flavonoids have hypoglycemic properties because they improve altered glucose and oxidative metabolisms of the diabetic states. They also exert a stimulatory effect on insulin secretion by changing Ca^{++} concentration (Song *et al.*, 2005). There are reports on the antidiabetic effect of flavonoids such as quercetin, isoquercetin, rutin, apigenin and naringenin (Coman *et al.*, 2012), which suggests the direct effect of flavonoids on insulin secretion on prevention of beta-cell apoptosis and modulation of proliferation of β–cells. (Pinent *et al.*, 2008). In view of the available scientific reports, flavonoids are known to possess high antioxidant activity and have been demonstrated to act on biological targets involved in type 2 diabetes mellitus such as: α-glucosidase, glucose transporter or aldose reductase (Nicolle *et al.*, 2011).

Further pharmacological and biochemical investigations has to be carried out to find out the active constituents responsible for antidiabetic activity and to elucidate its mechanism of action.



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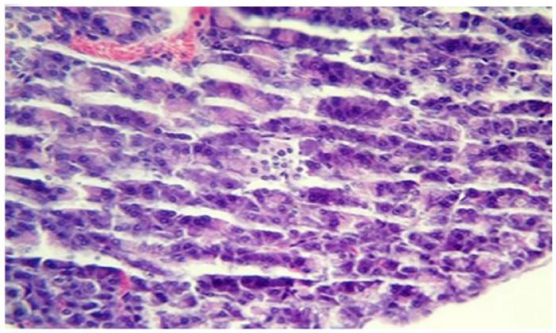
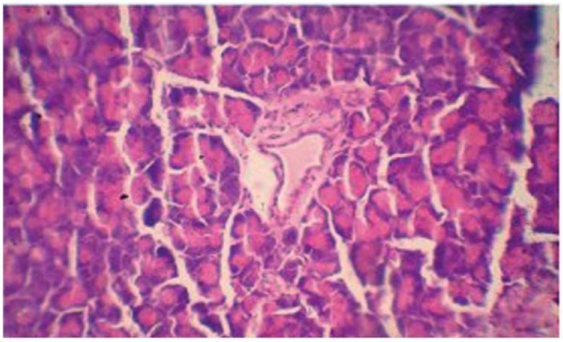
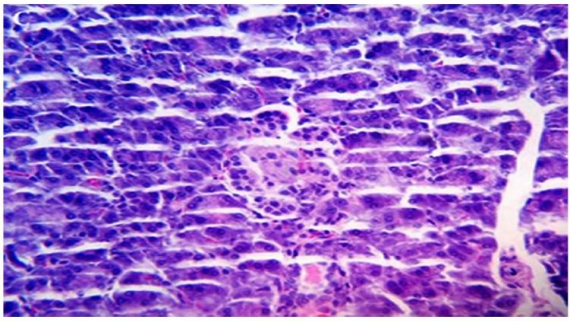


B

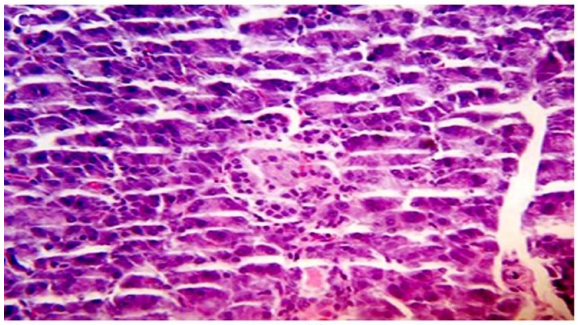
Plate II Histopathology of Pancreas in Normal and diabetic rats, 400X A- Normal rats, B- STZ induced diabetic rats



A



B



C

Plate III: Histopathology of pancreas in treatment groups, 400X
A- Glibenclamide treated group B- Methanolic extract of *F. montana* leaf @ 250 mg/kg C- Fruit juice of F.montana @ 2.5 ml/kg

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7. Ethical Matter
The above mentioned research work was part of the corresponding authors M.V.Sc. thesis work and laboratory research work was done as per IAEC norms with humane considerations.

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