Napata College
Pharmacy program

A thesis submitted by the faculty of Pharmacy of Napata college in partial fulfillment of the requirements of B.Sc. degree in pharmacy

The effects of salvia officinalis extract on modulating blood glucose levels in albino Rat

Prepared by:

Eman Mohamed Alamin Abbass

Israa Adel Amer Mohamed

Rula Ali Ahmed Mohamed

Rzaz Alamin Bashir nogod

zuhaira Mahmud Ali

Supervised by: Dr. Moutaz Al. Sheikh

Co-supervised by: Dr. Ali Awad Allah

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Our thanks also extended to all who have provided direct and indirect support to complete this study.

Dedication

To our families who have provide the time, efforts and emotional support to complete this study successfully. We sincerely appreciate it all.

Abstract

This study was carried out to investigate the hypoglycemic effects of aqueous and methanol extract of salvia officinalis in glucose induced hyperglycemic rat (*in vivo*).

Twenty rat were divided into four group each group contains five rats, each rat treated with glucose solution 1 g/kg body weight (glucose 50%) orally.

Group I consider as control group, group II treated with glibenclamide, group III was treated by aqueous extract, group IV was treated by methanol extract and the blood glucose level was evaluated in time 0,30,60,120and 240 and recorded after that the result was analyzed and comparative with control group and group treated by glibenclamide using glucometer.

During oral glucose tolerance test (OGTT) aqueous extract of s. Officinalis leaves at 500 mg/kg doses clearly reduced blood glucose level in albino rat with percentage decrease of 64% as compared to standard drug glibenclamide (5 mg/kg) with percentage decrease of 75%.

Methanol extraction 500mg/dl dose have the least percentage of decrease (32%) even less than control group 39%. These outcomes suggest that aqueous extract possess a hypoglycemic principle can be useful for the treatment of diabetes.

الخلاصة

أجريت هذه الدراسة للتحقيق من التأثيرات الخافضة للسكر في الدم للمستخلص المائي ومستخلص بواسطة الميثانول في نبات الميرمية باستخدام الجرذان التي تم زيادة سكر الدم بالجلوكوز (في الجسم الحي).

تم تقسيم عشرين فأرا إلى أربع مجموعات كل مجموعة تحتوي على خمسة فئران ، كل جرذ عولج بمحلول الجلوكوز 1 جم / كجم من وزن الجسم (جلوكوز 50٪) بالفم.

المجموعة الأولى تعتبر مجموعة ضبط ، المجموعة الثانية عولجت بالجليبينكلاميد ، المجموعة الثالثة عولجت بالمستخلص المائي ، المجموعة الرابعة عولجت بمستخلص الميثانول وتم تقييم مستوى الجلوكوز في الدم في الوقت 0,30,60,120,240 دقيقة بعد ذلك تم تحليل النتيجة وتسجيلها ومقارنتها مع مجموعة الضبط والمجموعة التي عولجت بالجليبينكلاميد باستخدام مقياس الجلوكوز.

خلال اختبار تحمل الجلوكوز بالفم (OGTT)المستخلص المائي لأوراقنبات الميرمية عند جرعة 500 ملجم / كجم ، أدت الجرعات إلى انخفاض واضح في مستوى الجلوكوز في دم الجرذان البيضاء بنسبة انخفاض 64 ٪ مقارنة بالدواء القياسي glibenclamide (5 مجم / كجم) بنسبة انخفاض 75٪.

مستخلص الميثانول 500 ملجم / ديسيلتر لها أقل نسبة انخفاض (32٪) حتى أقل من مجموعة الضبط 39٪. تشير هذه النتائج إلى أن المستخلص المائي يمتلك مواد لها تاثير خافض لسكر الدم يمكن أن يكون مفيدًا في علاج مرض السكري.

Acronyms

DM ---------- Diabetes Mellitus

CVD----------cardiovascular disease

T2DM----------Type 2 diabetes

PPHG---------- postprandial hyperglycemia

OGTT---------- Oral glucose tolerance test

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CHAPTER ONE
INTRODUCTION

1. Introduction:

1.1.Diabetes Mellitus

is a non-communicable metabolic disorder that is chronic where there is derangement of insulin action, secretion, or both. A lack of Insulin results in disturbed carbohydrate, protein, and fat metabolism. There are genetic and environmental factors at play for the development of Diabetes Mellitus. The decrease in insulin secretion, reduction in glucose utilization, or increased gluconeogenesis ultimately result in hyperglycemia and harmful changes in different organs. (1,2).

The general categories into which diabetes is classified are as follows: a) Type 1 diabetes b) Type 2 diabetes (c) Diabetes of specific types resulting from other causes like exocrine pancreas disease, chemical or drug-induced diabetes, monogenic diabetes syndrome (d) Gestational Diabetes Mellitus. (3) In the world at present, 422 million people are suffering from Diabetes Mellitus. Every year there are 1.6 million fatalities linked directly to diabetes. (4) As per data obtained in studies done throughout the world, an estimation has been made by the International Diabetes Federation that by the year 2045, there will be about 693 million cases of diabetes in the age range of 18–99 years. (5)

1.2. Alternative medication for diabetes mellitus:

Salvia spp. have been reported to produce various phenolic metabolites that have gained much attention in relevance to their antioxidant, antimicrobial, antidiabetic, neuroprotective, anti-inflammatory as well as cytotoxic properties. (6,7).

Generally,the utilization of therapeutic plants is expanding around the globe, coupled with the tremendous expansion of alternative medicine and growing demand in health treatment. Recently, there has been a grown interest in the use of medicinal plants and plant kingdom has become an interesting target for the search by multinational drug and biologically active lead compounds. Plants are applied in pharmaceuticals to preserve and expand health physically, mentally and as well as to treat particular health conditions and afflictions and Traditional medicines in developing countries become one of the choice for primary healthcare because of it better cultural acceptability, better compatibility with the human body and lesser side effects than modern therapies.

Salvia spp. Was acknowledged for its excellent medicinal benefits. The genus Salvia L. belongs to the Lamiaceae family and shows about 900 species dispersed worldwide, mainly in the areas of the Mediterranean, Southeast Africa, Central and South America. (8). The Salvia name comes from the Latin word “salvare” which means “to heal” and “to save” and

has been used as a traditional remedy against diabetes in many countries for its glucose-lowering effect (9)**.**

1.3. Complications related to diabetes

Complications related to diabetes are predicted to significantly impact the economy and society as the prevalence of the disease grows. (10-14) The acute complications of diabetes include ketoacidosis or severe hypoglycemia. Examples of chronic disease complications are retinopathy, neuropathy, cardiovascular disease, and nephropathy. (14-18)- especially type 2 diabetes, is an epidemic requiring global attention as a CVD risk. In addition to well-known microvascular complications such as retinopathy or nephropathy, diabetes confers the substantial burden of CVD morbidity and mortality through macrovascular complications even in early or

pre-stages. Because of its asymptomatic onset and progression, population-based screening is essential for early detection of diabetes mellitus before the development of vascular complications, including CVD. Many modifiable risk factors such as hyperglycemia, hypertension, or dyslipidemia must be adequately and simultaneously controlled for prevention of CVDs in people with established diabetes mellitus. (19).

If we could avoid this complication and decrease the mortality rate due to diabetes with natural, safe, effective, less side effect and less cost treatment we can take it to the next level.

CHAPTER TWO
LITERATURE REVIEW

2.litrature

2.1. literature Review

The available management strategies of diabetes are quite expensive and sometimes unsafe. This necessitates the need for bio-active drugs from medicinal plants. Although Salvia officinalis (sage) is used in herbal medicine, the scientific validation for anti-diabetic effects of various extracts have been elusive.

In previous study that aimed to determine and compare the anti-hyperglycemic efficacy of methanolic, hexane, ethyl acetate, and aqueous leaf extracts of Salvia officinalis in alloxan-induced diabetic mice. Phytochemical screening of the extracts revealed presence of flavanone, sterols, saponins, tannins, alkaloids, and triterpenes. The extracts were subjected to preliminary in vivo bio-assays at dosage levels of 400 mg/kg for 7 days through oral administration. The aqueous extract demonstrated significant hypoglycemic effect, $p<0.05$ hence subjected to further hypoglycemic studies for 15 days. There was a significant decrease in blood sugar levels of groups treated with aqueous extract at 400 mg/kg and 600 mg/kg doses from 452.00 ± 11.13 mg/dL and 431.00 ± 10.65 mg/dL to 256.33 ± 5.12 mg/dL and 256.67 ± 8.74 mg/dL. Weight gain improved significantly from 28.05 ± 0.39 g and 27.38 ± 0.52 g to 29.32 ± 0.42 g and 28.55 ± 0.38 g respectively compared to controls, $p<0.05$. Histopathological studies revealed no significant changes in liver and kidney tissues. Besides, no significant cytotoxic effect was reported. Results from this study indicate that aqueous extract of Salvia officinalis is a potential anti-hyperglycemic and can be used in modulating blood glucose levels. (20)

The evaluation of S. officinalis tea infusion showed comparable efficacy with metformin, which is normally used for type II diabetes treatment; Common sage (Salvia officinalis L.) is among the plants that are claimed to be beneficial to diabetic patients, and previous studies have suggested that some of its extracts have hypoglycemic effects on gluconeogenesis at the level of the liver. Primary cultures of hepatocytes from healthy, sage-tea-drinking rats showed, after stimulation, a high glucose uptake capacity and decreased gluconeogenesis in response to glucagon. Essential oil from sage further increased hepatocyte sensitivity to insulin and inhibited gluconeogenesis. Overall, these effects resemble those of the pharmaceutical drug metformin, a known inhibitor of gluconeogenesis used in the treatment and prevention of type 2 diabetes mellitus. However, its effects on fasting glucose levels in normal animals and its metformin-like effects on rat hepatocytes suggest that sage may be useful as a food supplement in the prevention of type 2 diabetes mellitus by lowering the plasma glucose of individuals at risk. (21).

Earlier studies have revealed the crucial role of phenolic components and flavonoids in both α-amylase and α-glucosidase inhibition. (22,23)

T2DM is a progressive disease characterized by a slow and continuous decline in β-cell function. α-Glucosidase and α-amylase are the enzymes involved in carbohydrate digestion, where their inhibition significantly reduces the postprandial increase of blood glucose. Hence, this serves as a strategic approach in blood glucose management among T2DM and borderline patients. (24). Hydrolysis of starch is possible by enzymes from amylases (endoamylases and exoamylases). The most well-known endoamylase is α-amylase, a calcium metalloenzyme that cannot function in the absence of calcium. α-amylase is present in both the salivary and pancreatic amylases of humans and certain plants, fungi and bacteria. αamylase inhibitors are also known as starch blockers that exert their antidiabetic mechanism by preventing starch absorption through blocking hydrolysis of 1,4-glycosidic linkages of starch and other oligosaccharides into maltose, maltotriose and other simple sugars. (25). The hydrolysis degree of the substrate by α-amylases can be divided into two categories, where saccharifying α-amylases hydrolyze 50–60% and liquefying α-amylases cleave about 30–40% of the glycosidic linkages of starch. α-Glucosidase is an essential enzyme in lysosomes for glucose degradation. (26). The enzyme inhibitors are capable of binding reversibly to the carbohydrate bindingregion of α-glucosidases. This bonding leads to competition with oligosaccharide binding and thus, delays oligosaccharides' cleavage into monosaccharides. These mechanisms highlight the action of α-glucosidase in retarding intestinal glucose absorption and PPHG. (27) . PPHG is one of the risk factors in diabetic patients that were identified to complicate T2DM treatment. Indirectly, α-glucosidase inhibitors play an important role in treating PPHG in diabetic patients. (28). Thus, the hydrolysis of oligosaccharides, trisaccharides, and disaccharides by membrane-bound intestinal α-glucosidase into glucose and other monosaccharides in the small intestine be will reduced, resulting in lower glucose circulation in the blood in T2DM.

known Salvia species particularly, (common sage) Salvia officinalis can be considered a relevant example of a plant with an evidence-based use in traditional medicine as well in modern phytotherapy. It is officialis recognized as a medicinal plant, with the leaves being used not only to prepare extracts for the treatment of skin disorders, minor wounds, mouth and throat disorders, but also in the treatment of gastrointestinal disorders in form of comminuted herb or aqueous and

hydro-alcoholic extracts for oral use (29). Chemical composition and relative variability of S. officinalis has been extensively investigated, and a plethora of factors have been recognized

as relevant for qualitative and quantitative composition, such as environmental, physiological and morphologic factors, genotypes, age, environmental stress and agronomic procedures, climatic conditions, season, salinity and culture site (30,31). The multipurpose applications of sage essential oil, derived from traditional use or as potential new approaches, including medicinal use, are largely supported by experimental phytochemical, biochemical and pharmacological studies as well as scientific reviews. (32,33). Polar fractions contain polyphenols, such as rosmarinic acid, ellagic acid, rutin, chlorogenic acid, quercetin and a lower amount of luteolin-7-glucoside, epicatechin, epigallocatechin gallate. Phenolic acids such as caffeic acid and 3-caffeoylquinic acid are also present. Aqueous and alcoholic extracts contain also several volatile components such as borneol, cineole, camphor and thujone. (34,35).

Sage is still one of the most common and widely used plants with numerous applications as aculinary herb mainly for the volatile aroma; as medicine due to the presence of polar and nonpolar biologically active metabolites and as cosmetic ingredient with traditional and innovative uses, for examples as anti-hyperhidrotic agent for deodorant formulations. Moreover, the less investigated Salvia species may represent a promising and unexplored field of research for innovative products with medicinal properties. (36)

To the best of our knowledge, only two scientific papers have focused on in-vivo study of salvia officinalis blood glucose lowering effect, one using alloxan-induced diabetic rats and the other using streptozotocin induced diabetes rats.

The present study aims to elucidate and compare blood glucose lowering effect of 2 different extract methanol and aqueous of salvia officinalis in-vivo using oral glucose tolerance test in glucose induced hyperglycemic rat.

The main goal of our research is to evaluate anti-hyperglycemic effect of S. officinalis, in order to support their potential health benefits.

CHAPTER THREE
MATERIALS AND METHODS

3. 1.MATERIALS AND METHODS

3.1.1. The plant materials

(s. officinalis) from local market during October 2022, The plant materials were identified and authenticated by botanists at the National Institute for Medicinal and Aromatic Plant Research, Khartoum Sudan.

3.1.2.**Animals**

Healthy adult albino rats of either sex weighing 70-270g were used for the study. They were obtained from Napata college animal house and faculty of veterinary Medicine-University of Khartoum. All the experimental procedures and protocols involving animals were ethical.

3.2. Methods

3.2.1. Extraction

Two diffrent extract was made: methanolic and aqueous.Extraction was carried out according to a method described by Sukhdevet. al. (2008). 100 g of plant sample was coarsely powdered using mortar and pestle and successively extracted with methanol using soxhlet extractor apparatus. Extraction carried out for about eight hours till the color of solvents at the last siphoning time returned colorless.

The aqueous extraction was carried by infusion method According to Radulescu, Chiliment, and Oprea (37), 250 mL of boiling water are poured over 100 g of leaves of Salvia officinalis L. and filtered after 30 min.

The two Solvents were evaporated and allowed to air in Petri dishes till complete dryness and the yield percentages were calculated as followed:

Weight of extract obtained / weight of plant sample X100. **(Table 1)**

3.3.2. Experimental protocol for *in vivo* hypoglycemic activity

Hypoglycemic effect in glucose induced hyperglycemic rat (OGTT)

Oral glucose tolerance test (OGTT) was performed according to the standard method (Xia et al., 2013) with minor modification.

4group ,5rat per group. Group I was treated as control group (Glucose solution 1 g/kg body weight), Group II was treated with glibenclamide (5 mg/kg body weight), Groups III, IV, were treated with Methanol and aqueous extract of S. officinalis leaves at 500 mg/kg body weight, respectively. Glucose solution (1 g/kg body weight) was administered at first. Then, drug and extract solutions were administered to the glucose fed. Serum glucose level of blood sample from tail vein was estimated using glucometer (on-call plus glucometer**,** PEF.G113-214**,** LOT:3367381**,**08.06.2022). (blood glucose test strip: LOT: 1692350.code:007**,** LOT: 1692370.code:336**,** LOT: 1692370.code:336**,** LOT: 1692016.code:311**,** LOT: 1692370.code336)at 0, 30, 60, 90, 120,240 min.

Percent decrease of blood glucose level after 240 min was measured using the following equation **(table 6).**

$$\text{Decrease (\%)} = \frac{\text{GL0 min} - \text{GL240 min}}{\text{GL0 min}} \times 100$$

$GL0\ min$= Blood glucose level at 0 min

$GL120\ min$=Blood glucose level at 240 min

3.3. statistical analysis

The results were statistically analyzed using Microsoft office 2016, word and excel 2016.

CHAPTER FOUR

RESULTS& DISCUTIONS

4.1.Results

Table No (1): shows the yield of extraction

Extraction	Weight	yield
Methanol	5.9	5.9%
Aqueous	1.93	1.93%

Table No(2):shows blood Glucose level mg/dl. Group I (control group)

Rat/time	0	30	60	120	240
Rat 1	127	55	73	73	72
Rat 2	99	165	134	139	132
Rat 3	279	121	270	111	90
Rat 4	144	107	111	95	102
Rat 5	162	112	147	104	99
Mean	162.2	125	147	104.4	99

Figure No (1) : show blood glucose concentration mg/dl decrease per time for group I rat no (1)

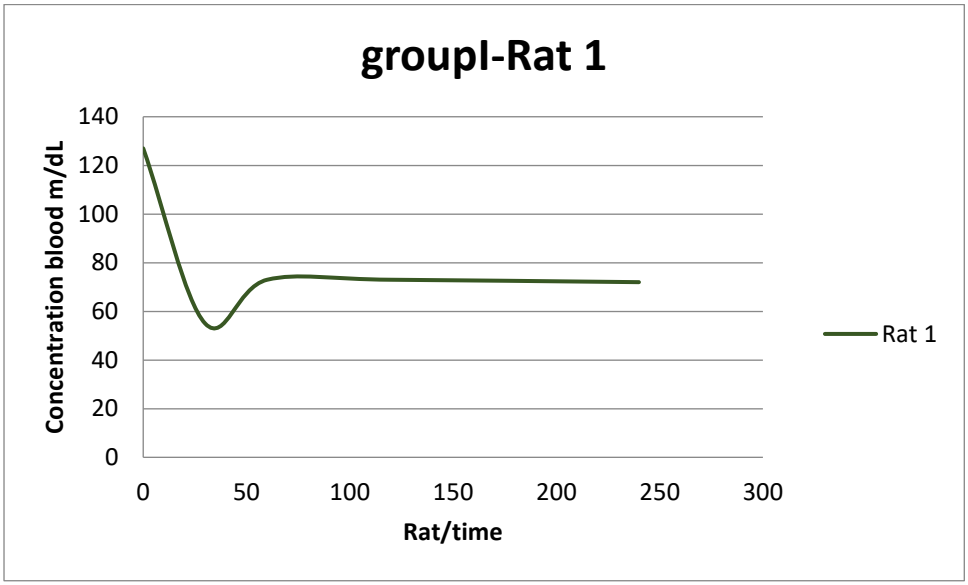


Figure No (2) : show blood glucose concentration mg/dl decrease per time for group I rat no (2)

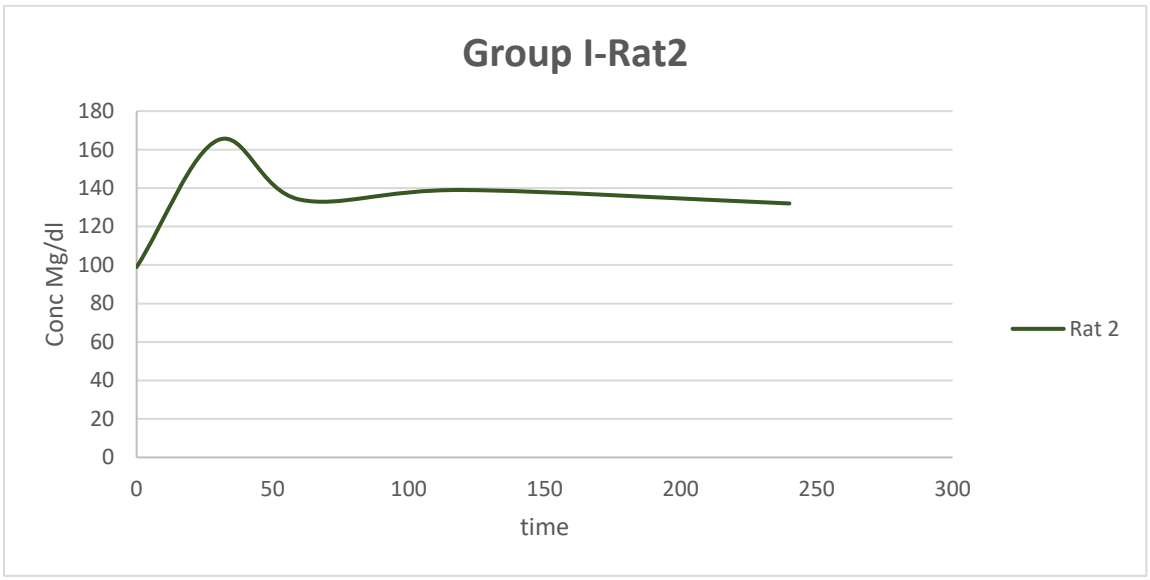


Figure No (3) : show blood glucose concentration mg/dl decrease per time for group I rat no (3)

Figure No (4) : show blood glucose concentration mg/dl decrease per time for group I rat no (4)

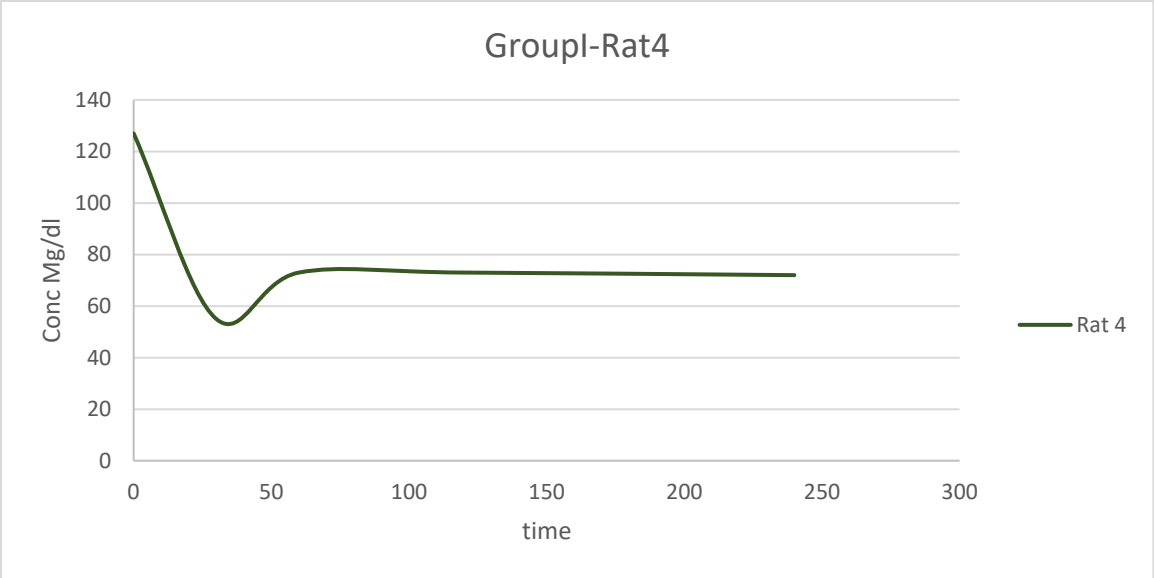


Figure No (5) : show blood glucose concentration mg/dl decrease per time for group I rat no (5)

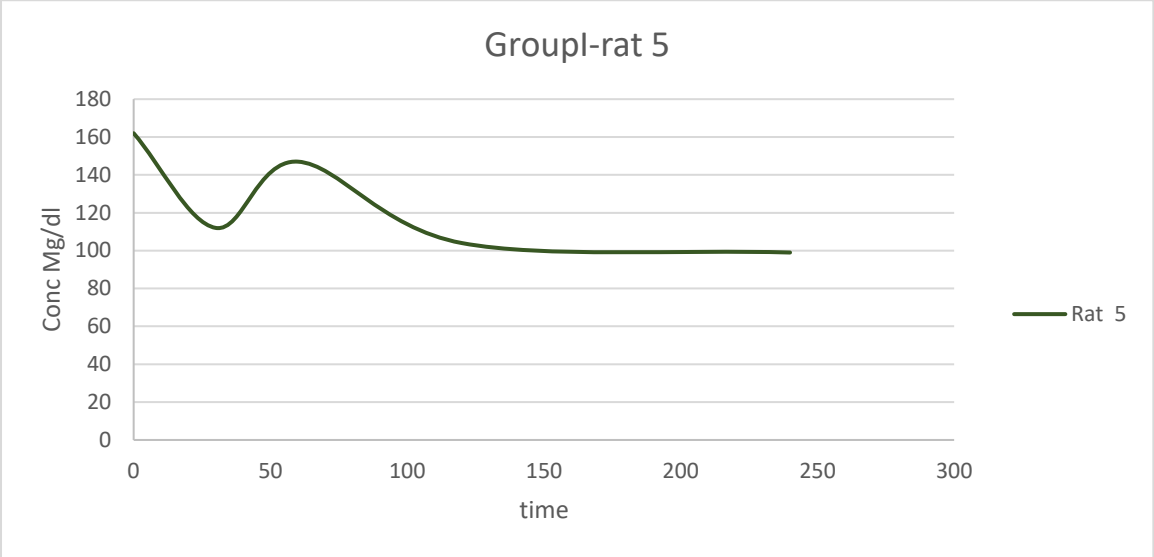


Figure No (6) : show blood glucose concentration mg/dl decrease per time for each rat in group I (mean).

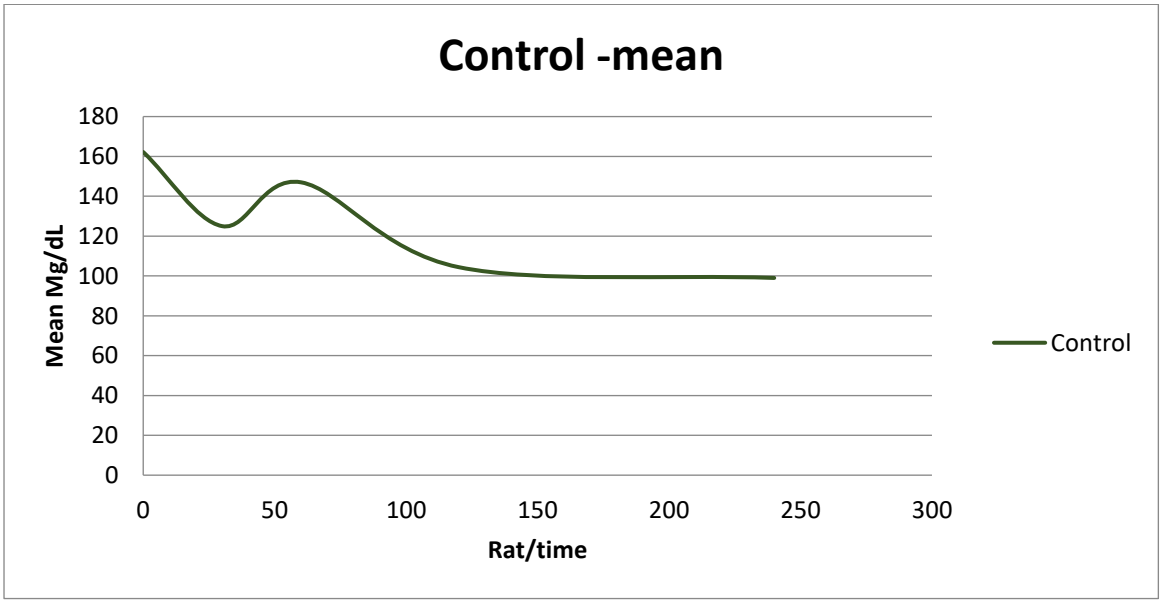


Table (3):shows blood Glucose level mg/dl. Group II, glibenclamide

Rat/time	**0**	**30**	**60**	**120**	**240**
Rat 1	104	98	104	104	57
Rat 2	227	134	101	113	113
Rat 3	134	116	126	58	72
Rat 4	509	227	89	59	53
Rat 5	480	131	104	67	55
Mean	290.8	141.2	104.8	80.2	70

Figure No (7) : show blood glucose concentration mg/dl decrease per time for group II rat no (1)

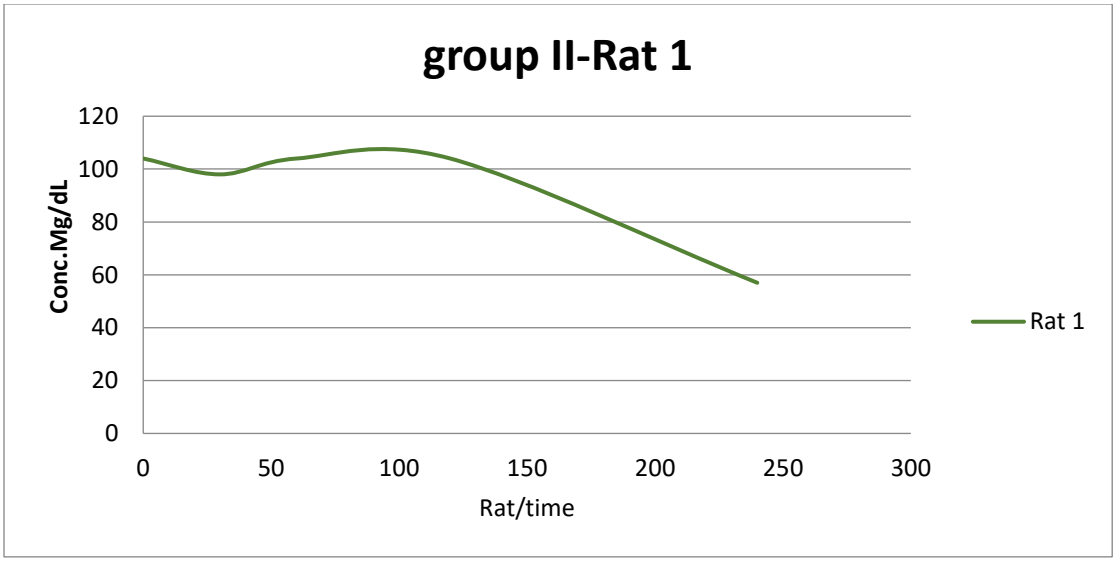


Figure No (8) : show blood glucose concentration mg/dl decrease per time for group II rat no (2)

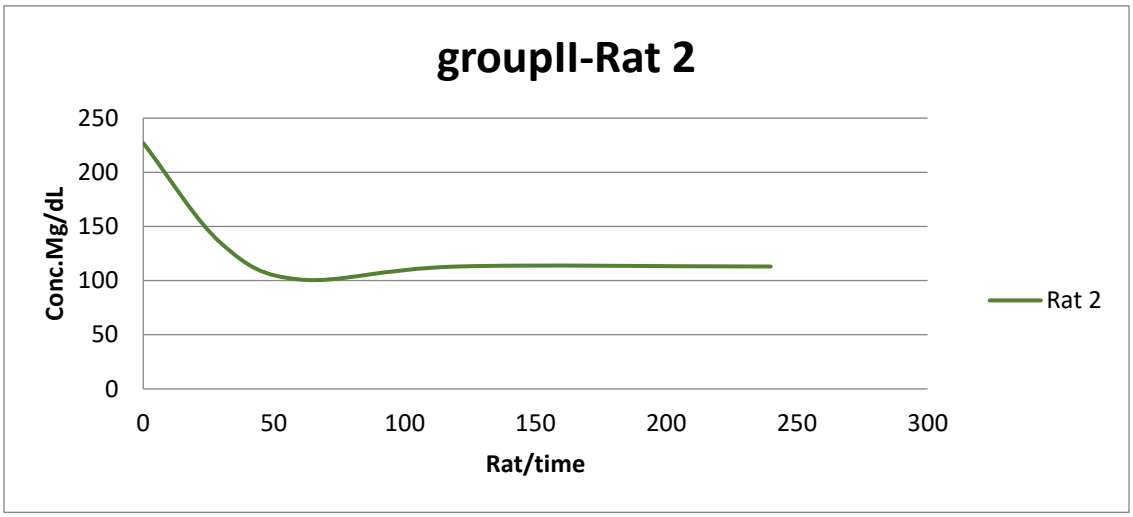


Figure No (9) : show blood glucose concentration mg/dl decrease per time for group II rat no (3)

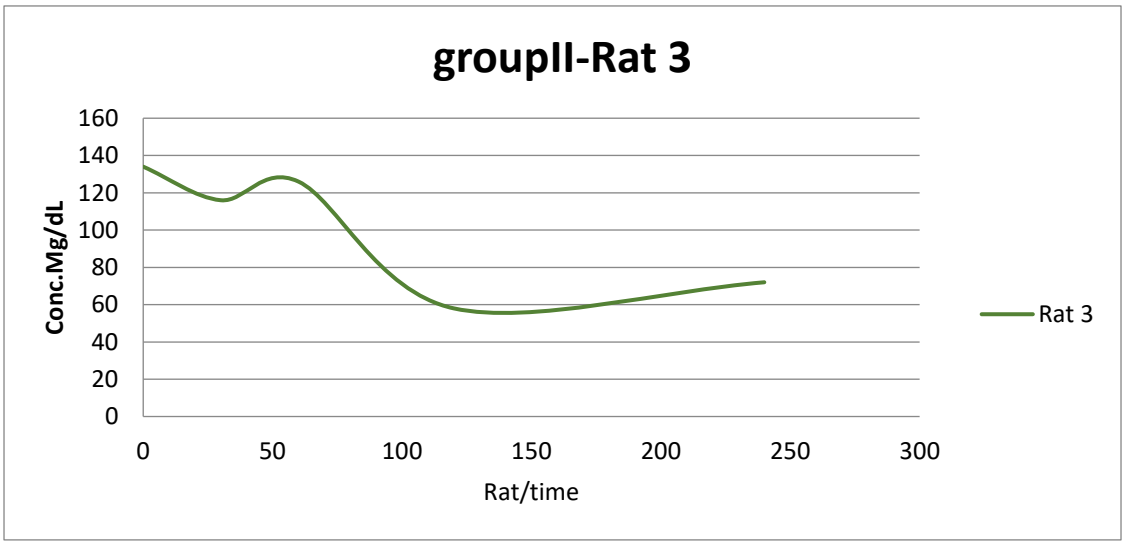


Figure No (10) : show blood glucose concentration mg/dl decrease per time for group II rat no (4)

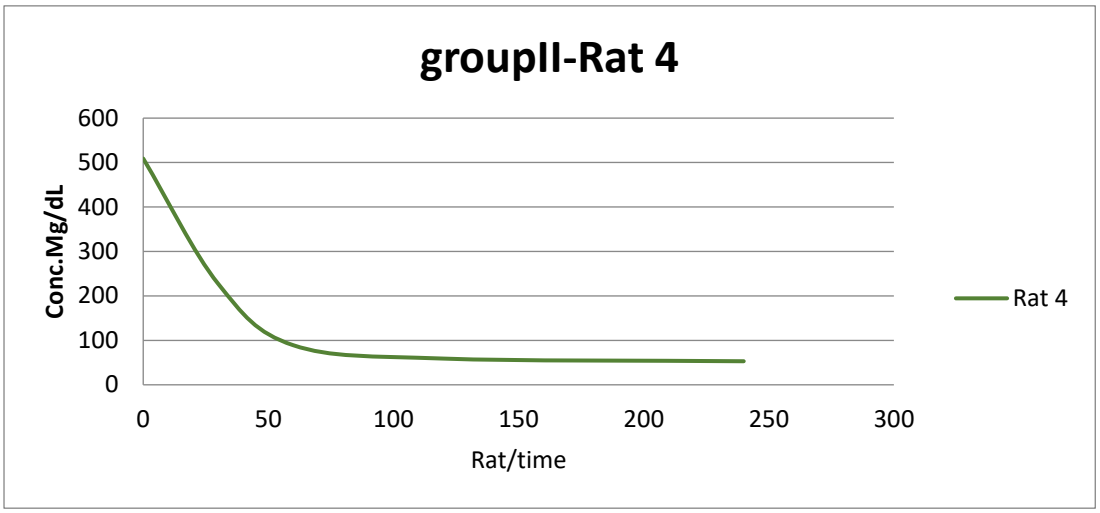


Figure No (11) : show blood glucose concentration mg/dl decrease per time for group II rat no (5)

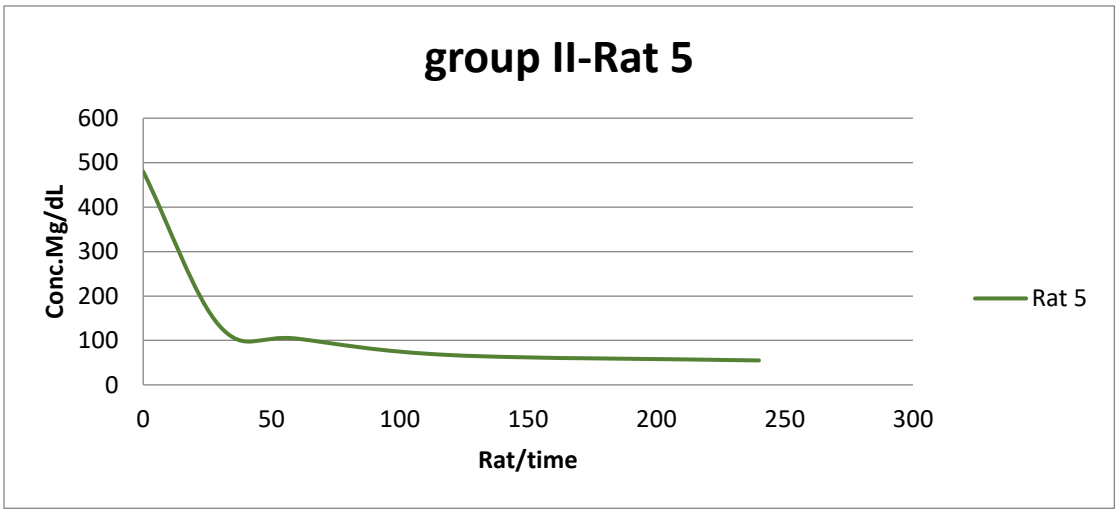


Figure No (12) : show blood glucose concentration mg/dl decrease per time for each rat in group II (mean).

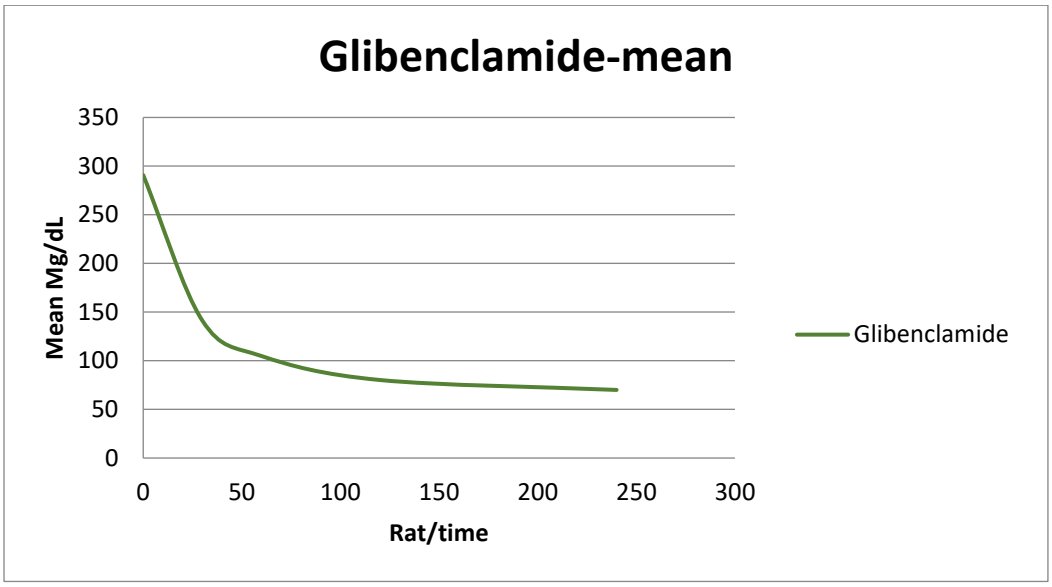


Table (4):shows blood Glucose level mg/dl. Groups III, Aqueous extract

Rat/time	**0**	**30**	**60**	**120**	**240**
Rat 1	345	193	80	70	66
Rat 2	81	93	69	64	62
Rat 3	117	128	108	99	89
Rat 4	102	79	73	49	56
Rat 5	260	62	85	53	53
Mean	181	111	82.6	67	65.2

Figure No (13) : show blood glucose concentration mg/dl decrease per time for group III rat no (1)

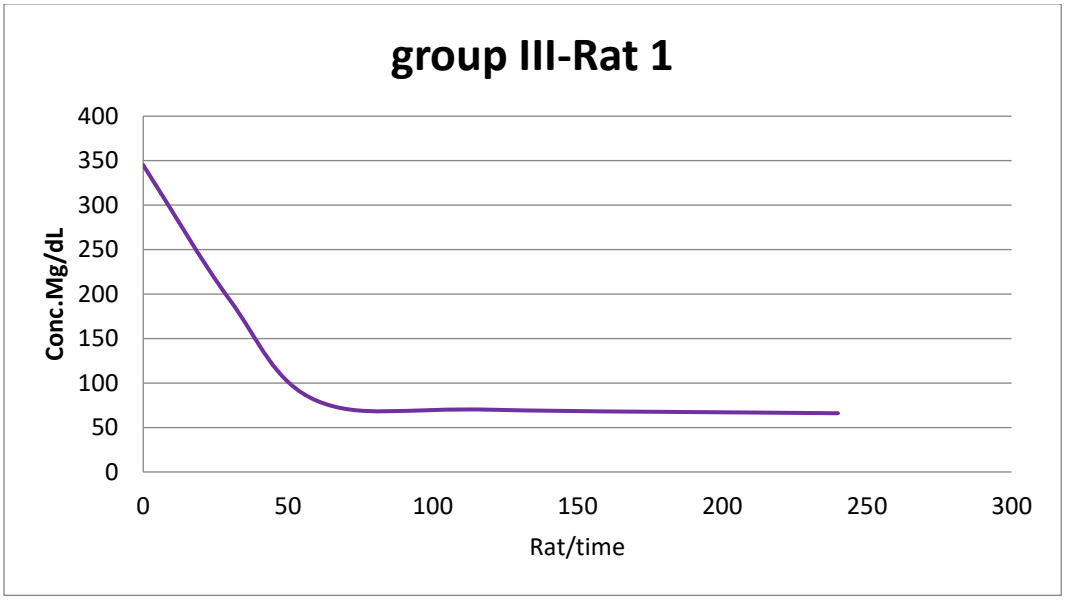


Figure No (14) : show blood glucose concentration mg/dl decrease per time for group III rat no (2)

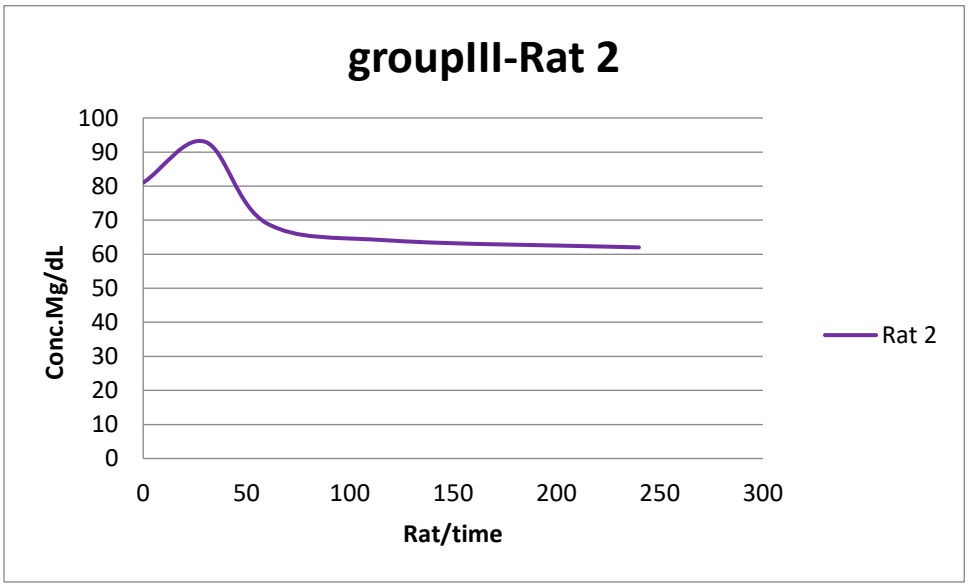


Figure No (15) : show blood glucose concentration mg/dl decrease per time for group III rat no (3)

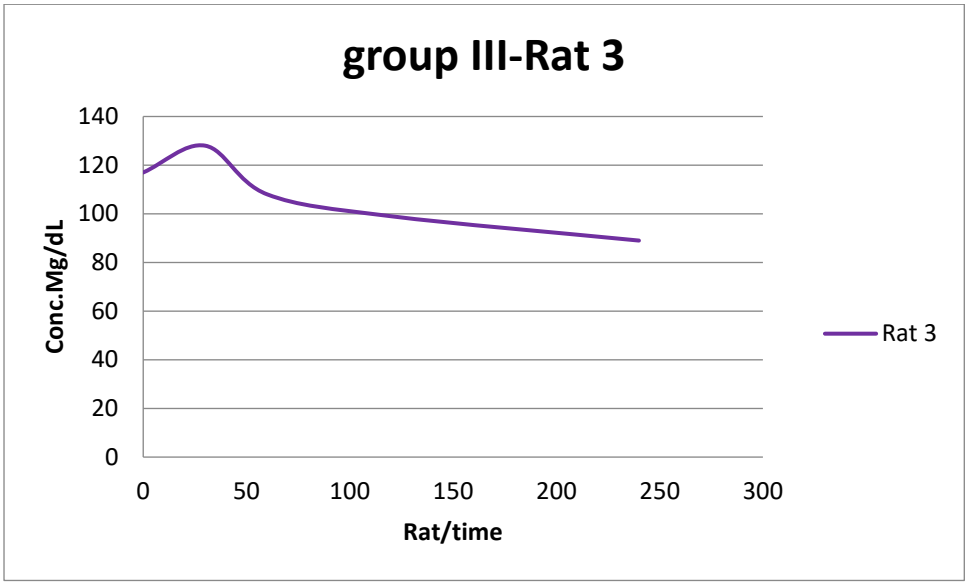


Figure No (16) : show blood glucose concentration mg/dl decrease per time for group III rat no (4)

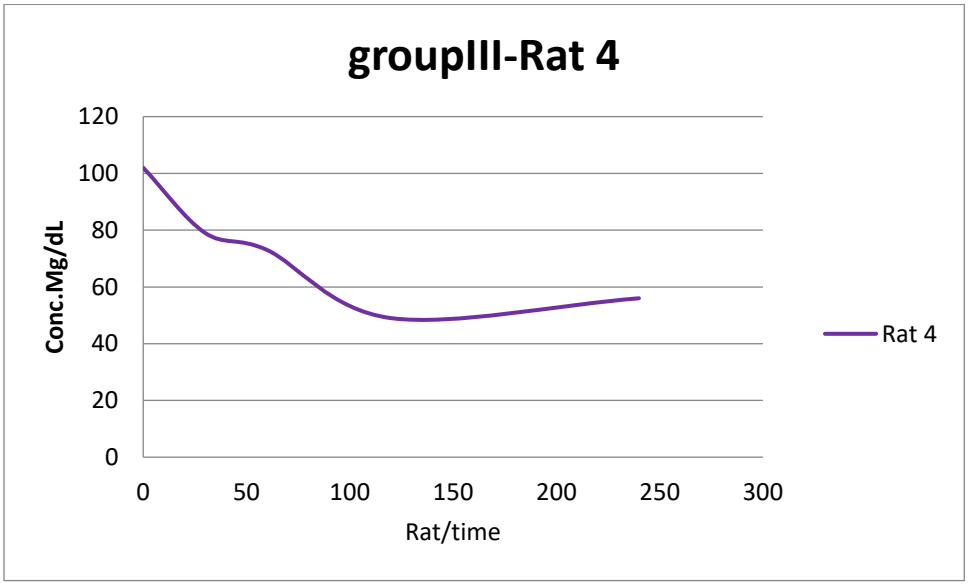


Figure No (17) : show blood glucose concentration mg/dl decrease per time for group III rat no (5)

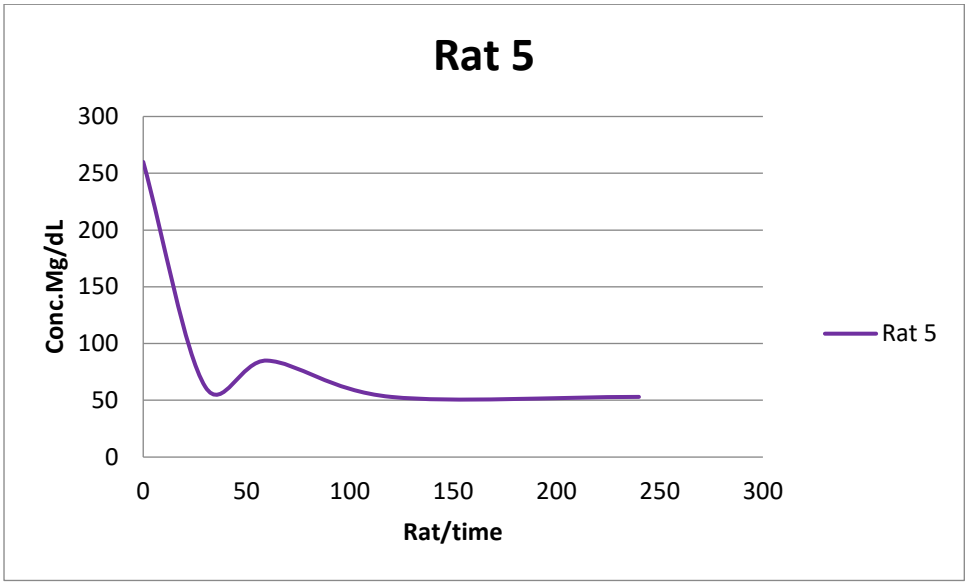


Figure No (18) : show blood glucose concentration mg/dl decrease per time for each rat in group III (mean).

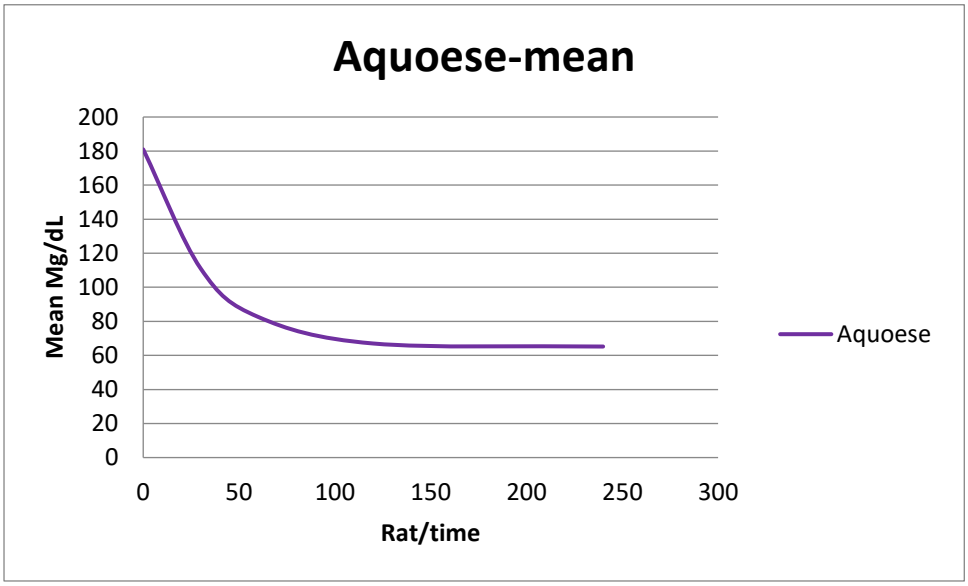


Table (5):shows blood Glucose level mg/dl. Group IV, Methanol extract

Rat/time	**0**	**30**	**60**	**120**	**240**
Rat 1	190	57	69	71	87
Rat 2	203	100	115	114	187
Rat 3	126	131	201	127	191
Rat 4	423	99	160	143	119
Rat 5	71	71	177	145	106
Mean	202.6	91.6	144.4	120	138

Figure No (19) : show blood glucose concentration mg/dl decrease per time for group IV rat no (1)

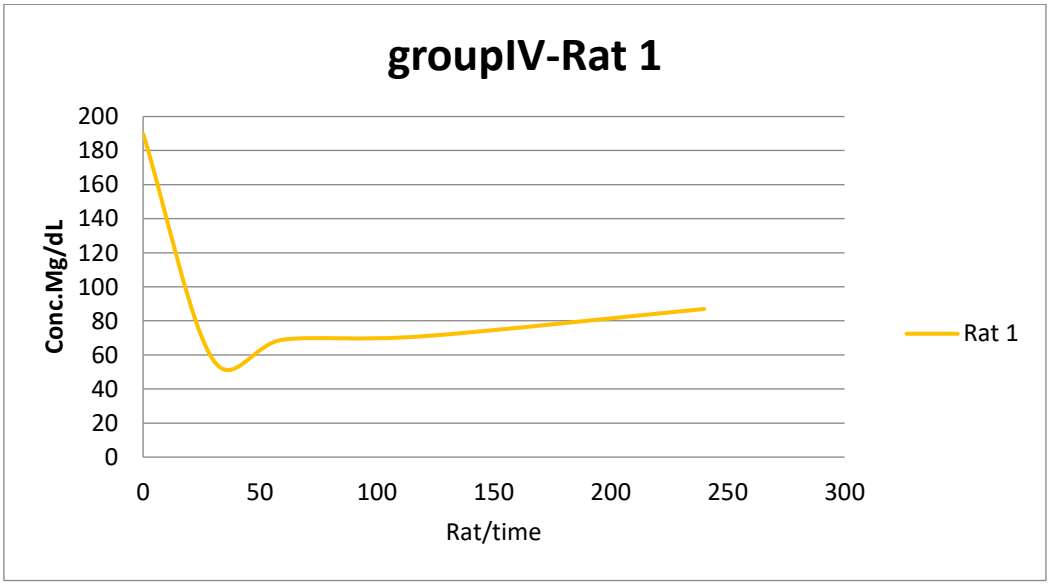


Figure No (20) : show blood glucose concentration mg/dl decrease per time for group IV rat no (2)

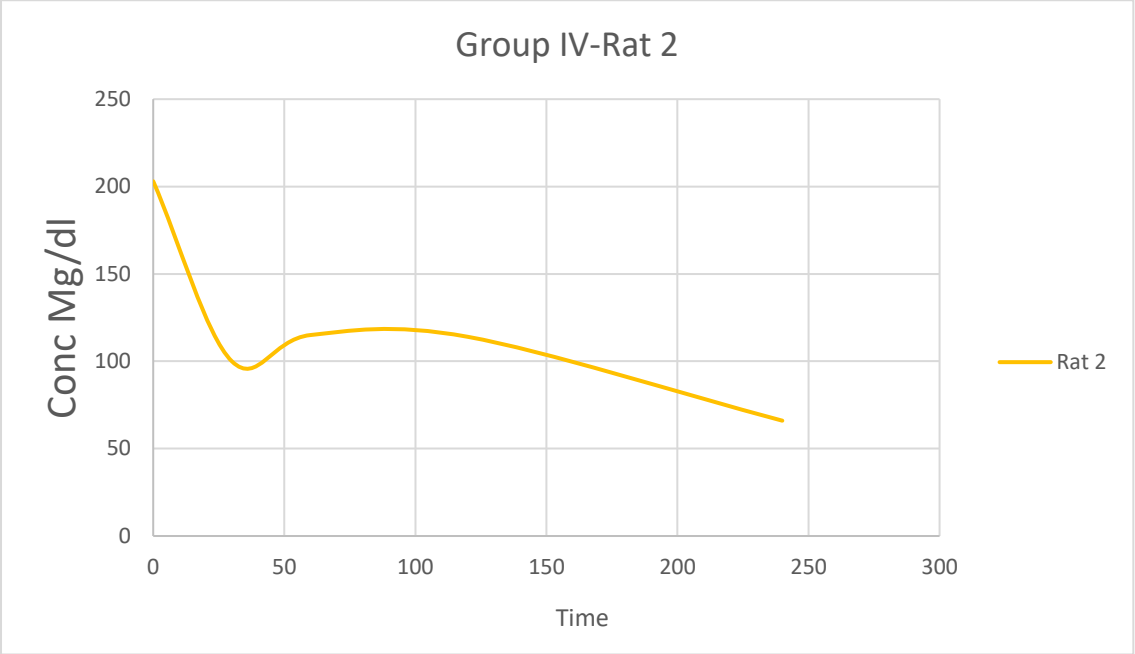


Figure No (21) : show blood glucose concentration mg/dl decrease per time for group IV rat no (3)

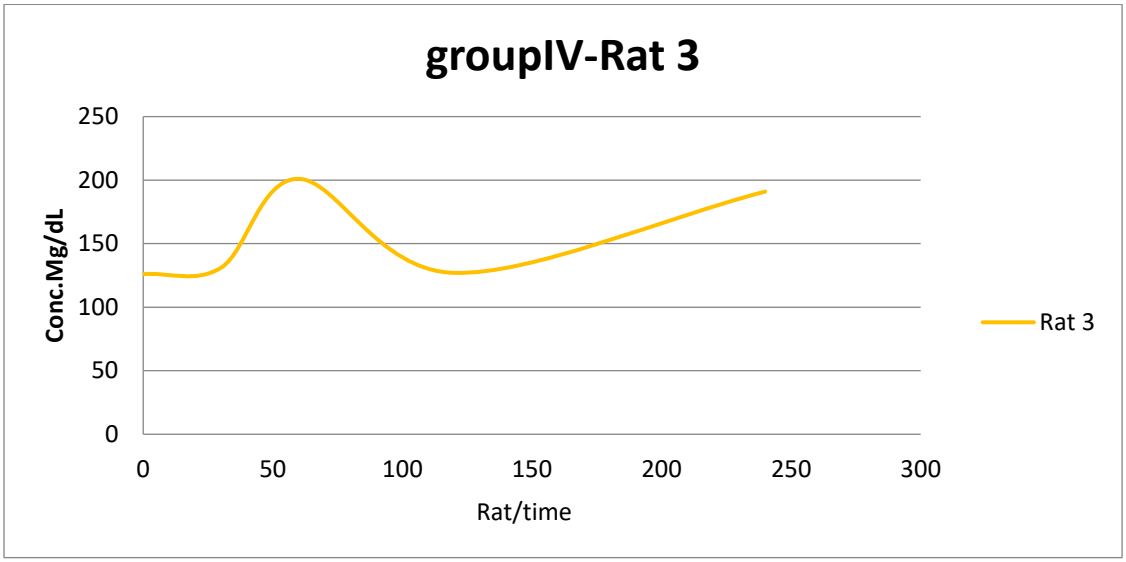


Figure No (22) : show blood glucose concentration mg/dl decrease per time for group IV rat no (4)

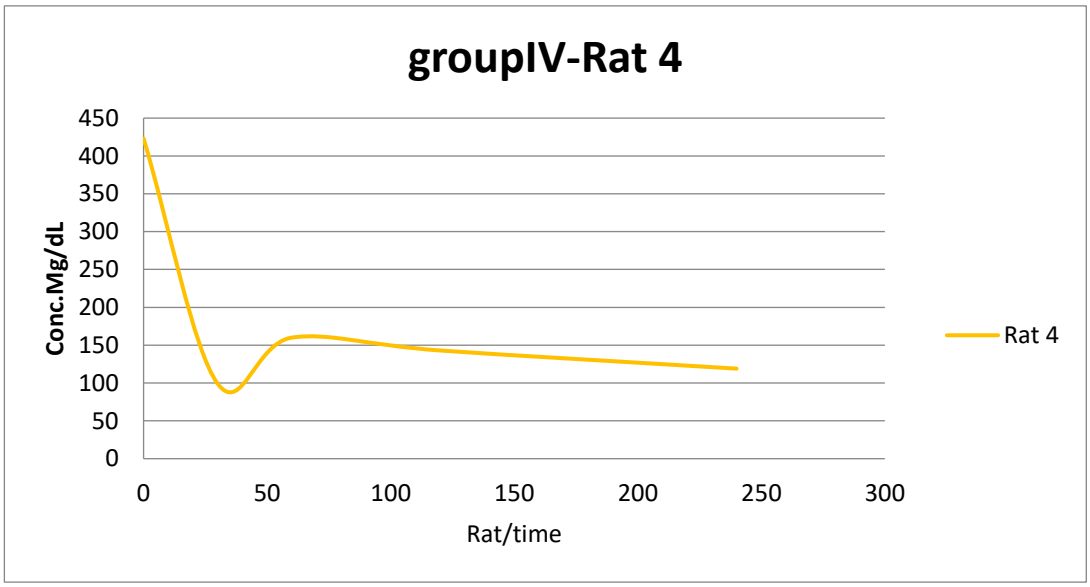


Figure No (23) : show blood glucose concentration mg/dl decrease per time for group IV rat no (5)

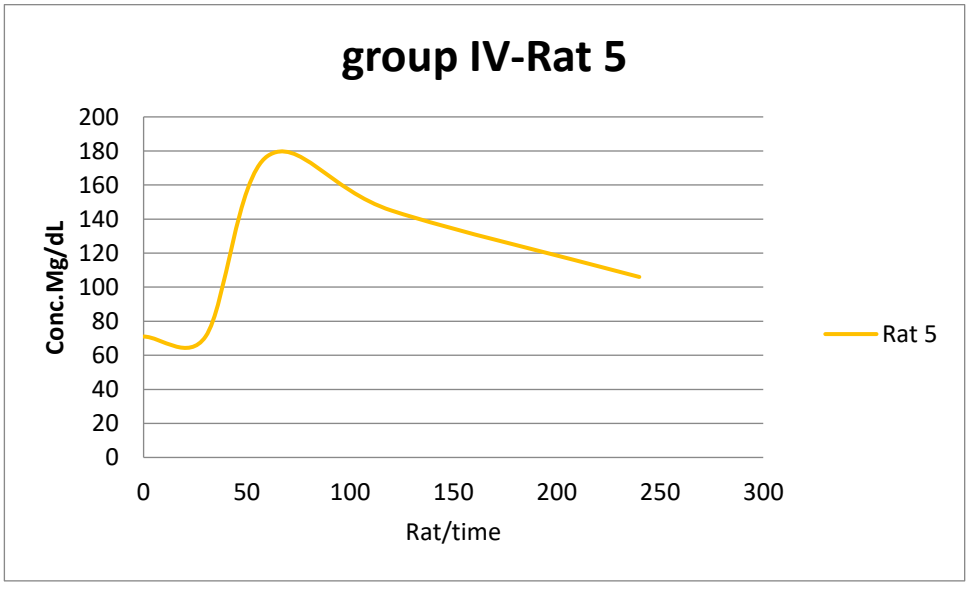


Figure No (24) : show blood glucose concentration mg/dl decrease per time for each rat in group IV (mean).

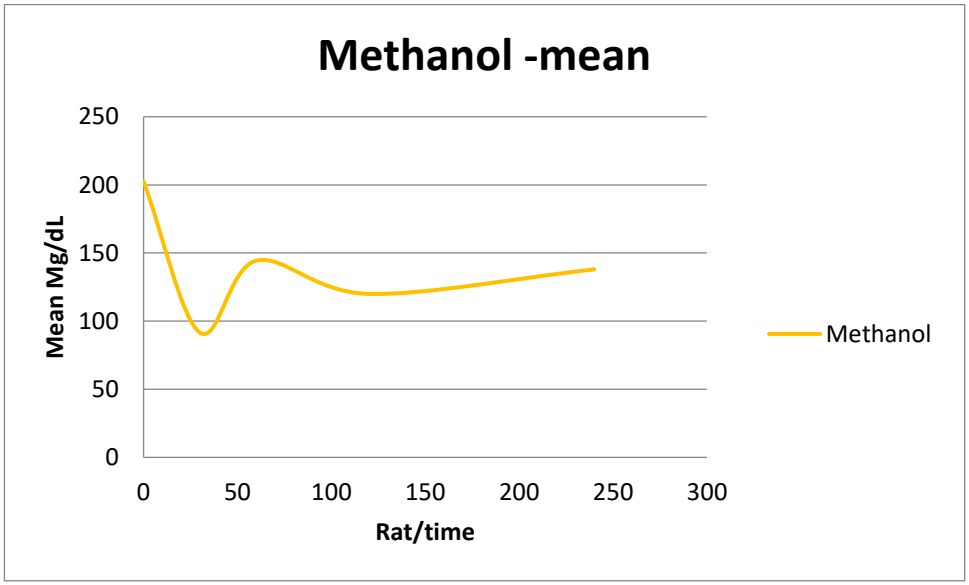


Figure No (25) : show blood glucose concentration mg/dl decrease per time for all groups

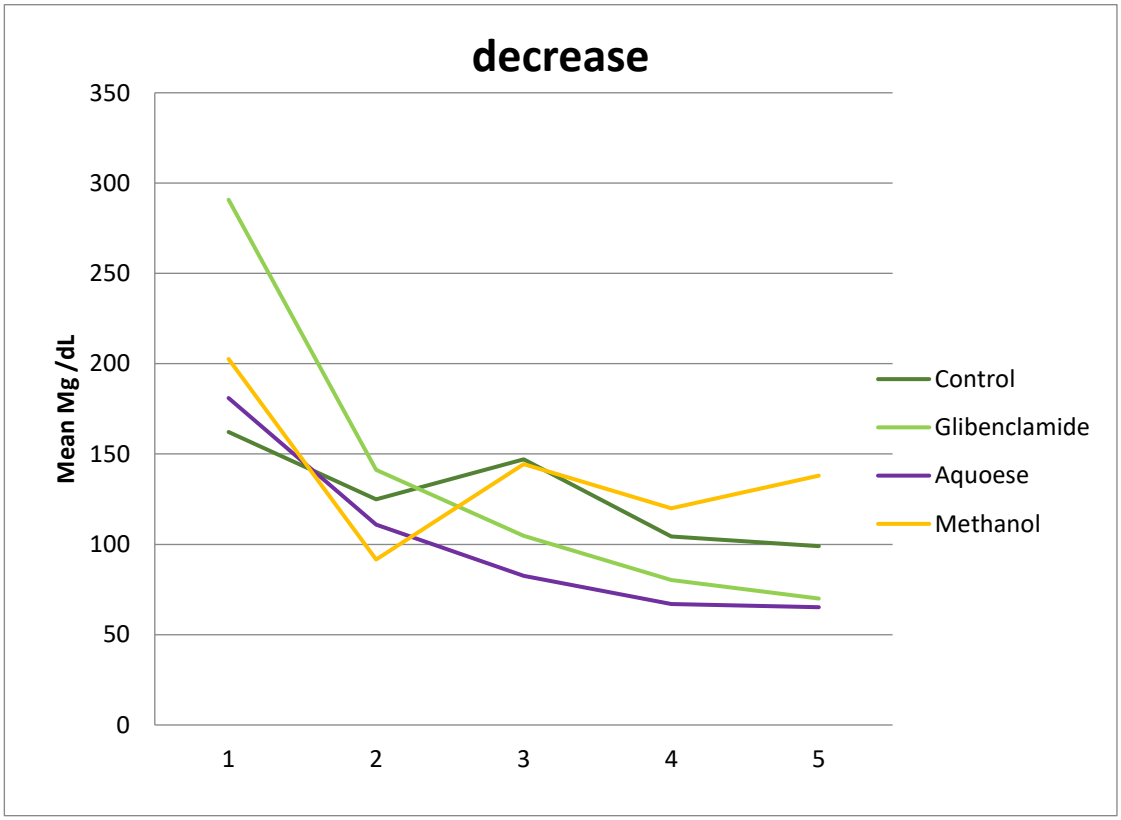


Table (6): shows Percent decrease of blood glucose level after 240 min

Group	Dose	Before (Mean)	Before (Mean)	Decrease(%)
Control	1g/kg body wt.	162	99	39%
Glibenclamide	5mg/kg Body wt.	291	70	76%
Aqueous	500mg/kg body wt.	181	65	64%
Methanol	500mg/kg body wt.	203	138	32%

Figure No (26) : show percentage decrease of blood glucose level after 240 mint

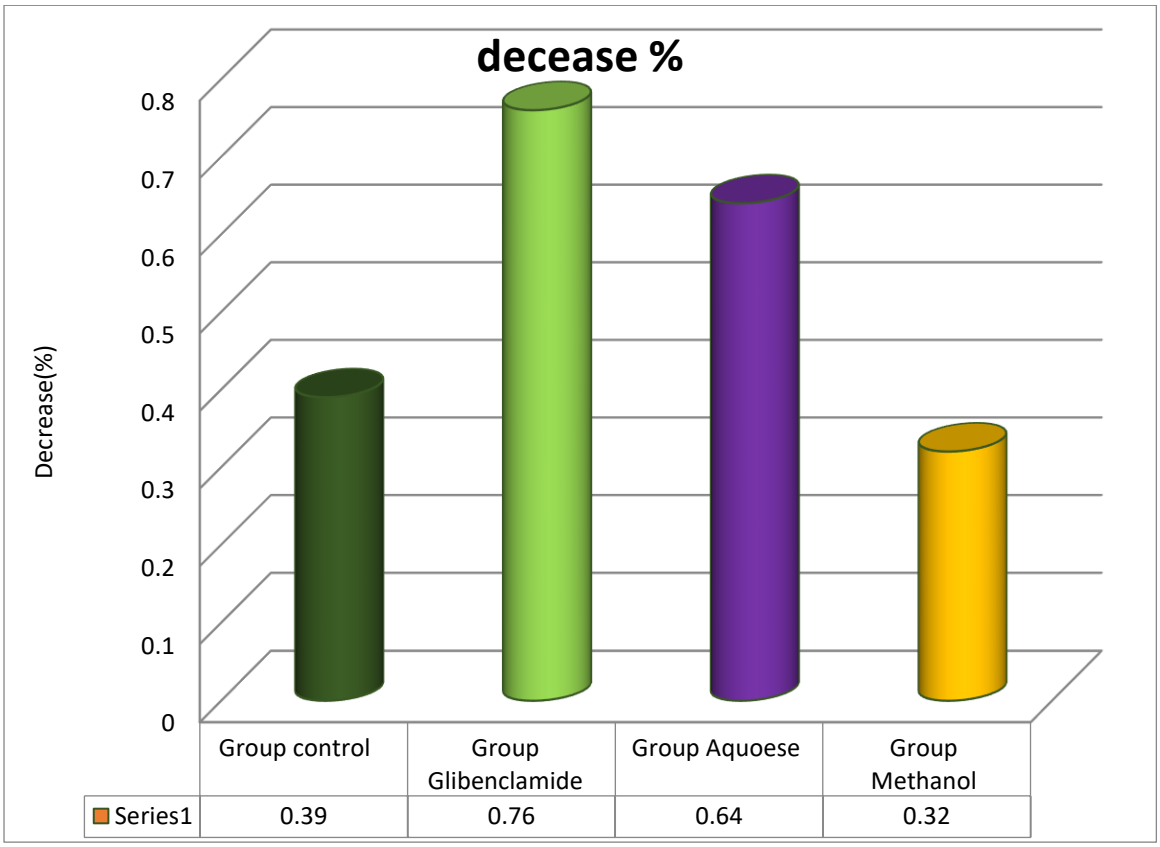


Table no. (7):Shows different between Rat control in glucose

Time	N	Mean	Std. Deviation	P-value
0	5	162.20	69.294	0.012
30	5	112.00	39.256	
60	5	147.00	74.280	
120	5	104.40	24.058	
240	5	99.00	21.840	
Total	**25**	**124.92**	**52.974**	

Table no. (8): Shows different between Rat Glibenclamide in glucose

Time	N	Mean	Std. Deviation	P-value
0	5	290.80	191.676	0.006
30	5	141.20	50.047	
60	5	104.80	13.368	
120	5	80.20	26.262	
240	5	70.00	25.179	
Total	**25**	**137.40**	**116.387**	

Table no .(9): Shows different between Rat Aquoesein glucose

Time	N	Mean	Std. Deviation	P-value
0	5	181.00	115.622	0.027
30	5	111.00	51.870	
60	5	83.00	15.281	
120	5	67.00	19.761	
240	5	65.20	14.237	
Total	**25**	**101.44**	**68.874**	

Table no. (10): Shows different between Rat Methanol in glucose

Time	N	Mean	Std. Deviation	P-value
0	5	202.60	134.091	0.014
30	5	91.60	28.719	
60	5	144.40	52.581	
120	5	120.00	30.166	
240	5	138.00	47.948	
Total	**25**	**139.32**	**74.310**	

4.2.Discussion:

In this study aqueous extract and methanolic extract of leaves of salvia officinalis exerted significant hypoglycemic activity in oral glucose tolerance test in glucose induced hyperglycemic rat.

Decrease of blood glucose level after 4hours of treatment is very well significant when compared with the control.

There are numerous pharmaceutical items which are accessible in current medicinal treatment have a long history of utilization as home grown cures, an expansive number of world's populace who live in creating nations cannot take the advantages of the present day pharmaceuticals as those are extremely expensive, so, research on phytomedicine has got great momentum in recent years to find out noble pharmaceuticals.

The OGTT is generally considered as more susceptible for the screening of impaired glycaemia, because it detects changes in post-prandial glycaemia that tend to precede changes in fasting glucose. All the current diagnostic criteria for diabetes depend on a threshold value imposed on a continuous distribution of blood glucose levels, yet the correct glycemic threshold that discriminates ‘normal’ from diabetic is not obvious. Though screening for undiagnosed type 2 diabetes remains a continuousissue, there is clear evidence that once it is diagnosed, complications can be prevented in many patients (Vamos et al., 2012; Hemmingsen et al., 2015). OGTT measures the body's ability to use glucose, the body’s main source (Kabir et al) of energy. OGTT can be used to diagnose pre diabetes and diabetes. The OGTT determines the shape of the glucose curve based on the measurements at 0, 30, 60, 90 and 120 and 240 min. The aqueous extract of S. officinalis showed significant ability to reduce the elevated glucose level in glucose induced hyperglycemic rat **Table (4)** it is ability to reduce the elevated glucose level is so closely to glibincalamid although it shows more hypoglycemic sings in subjected rats. And clearly have better lowering effect than methanol extract.

Methanolic extract showed a notable decrease in glucose level until time 60min, then it starts to exert hyperglycemic effect, this is due to either the bio-active compounds of the methanolic extract with hypoglycemic effect may have rabid execration time, or methanolic extract bio active compounds causing hyperglycemic effect start to take action after 60minutes (onset of action 60 min).

These outcomes suggest that aqueous extract possess a hypoglycemic principle with 64% percentage of decrease and can be useful for the treatment of diabetes. Further studies are warranted to isolate the active principle and to find out its accurate mechanism of action.

CHAPTER FIVE
CONCLUSIONS & RECOMMENDATIONS

5.1. Conclusion

From the study, it was concluded that S. officinalis may have hypoglycemic effect, but not sure about how this extracts can exert potent hypoglycemic activity although other suggested that some of its extracts have hypoglycemic effects on gluconeogenesis at the level of the liver and other suggest that it have its metformin-like effects on rat. It is a logical inference that this plant may recover the metabolism of glucose and increase insulin secretion by stimulating beta cells. It is possible to propose that the bioactive compounds present in the leaves extract and its fractions may be responsible for versatile effects. Based on the literature search.

5.2. Recommendation

- Further co-ordinate and well-structured studies would be required to isolate the bioactive compounds and determine their underlying molecular mechanism of action on diabetes induced rat model.
- These findings suggest that the plant may be a potential source for the development of new oral hypoglycemic agent.
- Further toxicity study are highly recommended .

5.3. CONFLICT OF INTEREST

The authors have not declared any conflict of interests

Reference:

1. American Diabetes Association. 2. Classification and diagnosis of diabetes: standards of medical care in diabetes-2021. Diabetes Care. 2021;44(Suppl 1): S15–S33. doi: 10.2337/dc21-S002 .
2. Asghari, B.; Salehi, P.; Sonboli, A.; Nejad Ebrahimi, S. Flavonoids from Salvia chloroleuca with α-Amylsae and α-Glucosidase Inhibitory Effect. Iran. J. Pharm. Res. 2015, 14, 609–615.8. Moradi-Afrapoli, F.; Asghari, B.; Saeidnia, S.; Ajani, Y.; Mirjani, M.; Malmir, M.; Dolatabadi Bazaz, R.; Hadjiakhoondi, A.; Salehi, P.; Hamburger, M.; et al. In vitro α-glucosidase inhibitory activity of phenolic constituents from aerial parts of Polygonum hyrcanicum. DARU J. Pharm. Sci. 2012, 20, 37.
3. Aung-Htut, M.T.; Ham, K.A.; Tchan, M.C.; Fletcher, S.; Wilton, S.D. Novel Mutations Found in Individuals with Adult-Onset Pompe Disease. Genes 2020, 11, 135.
4. Aynalem SB, Zeleke AJ. Prevalence of diabetes mellitus and its risk factors among individuals aged 15 years and above in Mizan-Aman Town, Southwest Ethiopia, 2016: a Cross-Sectional Study. Int JEndocrinol.2018;2018:9317987.doi:10.1155/2018/9317987.
5. Bhupathiraju SN, Hu FB. Epidemiology of obesity and diabetes and their cardiovascular complications. Circ Res. 2016;118(11):1723–1735. doi: 10.1161/CIRCRESAHA.115.306825
6. Blaslov K, Naranđa FS, Kruljac I, Renar IP. Treatment approach to type 2 diabetes: past, present, and future. World J Diabetes.2018;9(12):209219.doi:10.4239/wjd.v9.i12.209.
7. Cho NH, Shaw JE, Karuranga S, et al. IDF diabetes atlas: global estimates of diabetes prevalence for 2017 and projections for 2045. Diabetes Res Clin Pract. 2018; 138:271–281. doi: 10.1016/j.diabres.2018.02.023 .
8. Dall TM, Yang W, Halder P, et al. The economic burden of elevated blood glucose levels in 2012: diagnosed and undiagnosed diabetes, gestational diabetes mellitus, and prediabetes. Diabetes Care. 2014;37(12):3172–3179. doi: 10.2337/dc14-1036.
9. Dineshkumar, B.; Mitra, A.; Manjunatha, M. A comparative study of alpha amylase inhibitory activities of common anti-diabetic plants at Kharagpur 1 block. Int. J. Green Pharm. 2010, 4, 115–121.

10. Final European Union Herbal Monograph on Salvia officinalis L., Folium. Available online:ema.europa.eu/en/documents/herbal-monograph/final-european-union-herbal-monograph-salvia-officinalisl-folium-revision-1_en.pdf (accessed on 5 April 2020).
11. Fu, Z.; Wang, H.; Hu, X.; Sun, Z.; Han, C. The pharmacological properties of Salvia essential oils. J. Appl.Pharm. Sci. 2013, 3, 122–127.
12. Ghorbani, A.; Esmaeilizadeh, M. Pharmacological properties of Salvia o_cinalis and its components. J. Tradit.Complement. Med. 2017, 7, 433–440.
13. Grdiša, M.; Jug-Dujakovi´c, M.; Lonˇcari´c, M.; Carovi´c-Stanko, K.; Ninˇcevi´c, T.; Liber, Z.; Radosavljevi´c, I.;Šatovi´c, Z. Dalmatian sage (Salvia o_cinalis L.): A review of biochemical contents, medical properties andgenetic diversity. Agric. Conspec. Sci. 2015, 80, 69–78.
14. H, Sone.in Encyclopedia of Cardiovascular Research and Medicine, 2018.
15. H, Sone.in Encyclopedia of Cardiovascular Research and Medicine, 2018.
16. Hamidpour, M.; Hamidpour, R.; Hamidpour, S.; Shahlari, M. Chemistry, pharmacology, and medicinal property of sage (Salvia) to prevent and cure illnesses such as obesity, diabetes, depression, dementia, lupus,autism, heart disease, and cancer. J. Tradit. Complement. Med. 2014, 4, 82–88.
17. Hap K, Biernat K, Konieczny G. Patients with diabetes complicated by peripheral artery disease: the current state of knowledge on physiotherapy interventions. J Diabetes Res. 2021; 2021:5122494. doi: 10.1155/2021/5122494 .
18. Hap K, Biernat K, Konieczny G. Patients with diabetes complicated by peripheral artery disease: the current state of knowledge on physiotherapy interventions. J Diabetes Res. 2021; 2021:5122494. doi: 10.1155/2021/5122494.
19. Kanana Faith Mbiti, Maina Charles Mwendia, Kibet Joseph Mutai and Clement Josphat Matasyoh.Department of Biochemistry and Molecular Biology.Hypoglycaemic effects of Salvia officinalis extracts on alloxan-induced diabetic Swiss albino mice. Faculty of Science, Egerton University, P. O. Box 536 Egerton, Kenya. Department of Veterinary Parasitology, Microbiology and, Pathology, Faculty of Veterinary Medicine and Surgery, Egerton University, P. O. Box 536 Egerton, Kenya. Department of Chemistry, Faculty of Science, Egerton University, P. O. Box 536 Egerton, Kenya. 26 September, 2019.
20. Lebovitz, H.E. ALPHA-GLUCOSIDASE INHIBITORS. Endocrinol. Metab. Clin. N. Am. 1997, 26, 539–551.

21. Longaray Delamare, A.P.; Moschen-Pistorello, I.T.; Artico, L.; Atti-Serafini, L.; Echeverrigaray, S. Antibacterial activity of the essential oils of Salvia officinalis L. and Salvia triloba L. cultivated in South Brazil. Food Chem. 2007, 100, 603–608.
22. Lu, Y.; Foo, L.Y. Polyphenolics of Salvia-a review. Phytochemistry 2002, 59, 117–140.
23. Lu, Y.; Yeap Foo, L. Polyphenolics of Salvia—A review. Phytochemistry 2002, 59, 117–140
24. Martins N., Barros L., Sants-Buelga C., Henriques M.,Silva S.,Ferreira I.C.FR.Evaluation of bioactive properties and phenolic compound in different extracts prepared from salvia officinalis L.Food chem.2015;170:378-385.doi:10.1016/j.Foodchem.2014.08.096.
25. Maugeri G, Bucolo C, Drago F, et al. Attenuation of high glucose-induced damage in RPE cells through p38 MAPK signaling pathway inhibition. Front Pharmacol. 2021; 12:684680. doi: 10.3389/fphar.2021.684680.
26. Maugeri G, Bucolo C, Drago F, et al. Attenuation of high glucose-induced damage in RPE cells through p38 MAPK signaling pathway inhibition. Front Pharmacol. 2021; 12:684680. doi: 10.3389/fphar.2021.684680.
27. Menghini, L.; Leporini, L.; Pintore, G.; Chessa, M.; Tirillini, B. Essential oil content and composition of three sage varieties grown in Central Italy. J. Med. Plants Res. 2013, 7, 480–489.
28. Metformin-like effect of Salvia officinalis (common sage): is it useful in diabetes prevention? Cristovao F. Lima, Marisa F. Azevedo, Rita Araujo, Manuel Fernandes-Ferreira and Cristina Pereira-Wilson*Department of Biology, Centre of Biology, School of Sciences, University of Minho, 4710-057 Braga, Portugal; British Journal of Nutrition (2006).
29. Metformin-like effect of Salvia officinalis (common sage): is it useful in diabetes prevention? Cristovao F. Lima, Marisa F. Azevedo, Rita Araujo, Manuel Fernandes-Ferreira and Cristina Pereira-Wilson*Department of Biology, Centre of Biology, School of Sciences, University of Minho, 4710-057 Braga, Portugal; British Journal of Nutrition (2006).
30. Moradi-Afrapoli, F.; Asghari, B.; Saeidnia, S.; Ajani, Y.; Mirjani, M.; Malmir, M.; Dolatabadi Bazaz, R.; Hadjiakhoondi, A.; Salehi, P.; Hamburger, M.; et al. In vitro α-glucosidase inhibitory activity of phenolic constituents from aerial parts of Polygonum hyrcanicum. DARU J. Pharm. Sci. 2012, 20, 37.

31. Nickerson HD, Dutta S. Diabetic complications: current challenges and opportunities. J Cardiovasc Transl Res. 2012;5(4):375–379. doi: 10.1007/s12265-012-9388-1.
32. Nickerson HD, Dutta S. Diabetic complications: current challenges and opportunities. J Cardiovasc Transl Res. 2012;5(4):375–379. doi: 10.1007/s12265-012-9388-1.
33. Notkins, A.L. Immunologic and genetic factors in type 1 diabetes. J. Biol. Chem. 2002, 277, 43545–43548.
34. Pettus JH, Zhou FL, Shepherd L, et al. Incidences of severe hypoglycemia and diabetic ketoacidosis and prevalence of microvascular complications stratified by age and glycemic control in U.S. adult patients with type 1 diabetes: a Real-World Study. Diabetes Care. 2019;42(12):2220–2227. doi: 10.2337/dc19-0830.
35. Pettus JH, Zhou FL, Shepherd L, et al. Incidences of severe hypoglycemia and diabetic ketoacidosis and prevalence of microvascular complications stratified by age and glycemic control in U.S. adult patients with type 1 diabetes: a Real-World Study. Diabetes Care. 2019;42(12):2220–2227. doi: 10.2337/dc19-0830.
36. Sarwar N, Gao P, Seshasai SR, Gobin R, Kaptoge S; Emerging Risk Factors Collaboration. Diabetes mellitus, fasting blood glucose concentration, and risk of vascular disease: a collaborative meta-analysis of 102 prospective studies. Lancet. 2010;375(9733):2215–2222. doi: 10.1016/S0140-6736(10)60484-9.
37. Seng JJB, Kwan YH, Lee VSY, et al. Differential health care use, diabetes-related complications, and mortality among five unique classes of patients with type 2 diabetes in Singapore: a latent class analysis of 71,125 patients. Diabetes Care. 2020;43(5):1048–1056. doi: 10.2337/dc19-2519.
38. Topçu, G. Bioactive triterpenoids from Salvia species. J. Nat. Prod. 2006, 69, 482–487.
39. Tundis, R.; Loizzo, M.R.; Menichini, F. Natural products as alpha-amylase and alpha-glucosidase inhibitors and their hypoglycaemic potential in the treatment of diabetes: An update. Mini Rev. Med. Chem. 2010, 10, 315–331.
40. United Kingdom Prospective Diabetes Study Group. United Kingdom Prospective Diabetes Study 24: A 6-year, randomized,controlled trial comparing sulfonylurea, insulin, and metformin therapy in patients with newly diagnosed type 2 diabetes thatcould not be controlled with diet therapy. United Kingdom Prospective Diabetes Study Group. Ann. Intern. Med. 1998, 128,165–175.

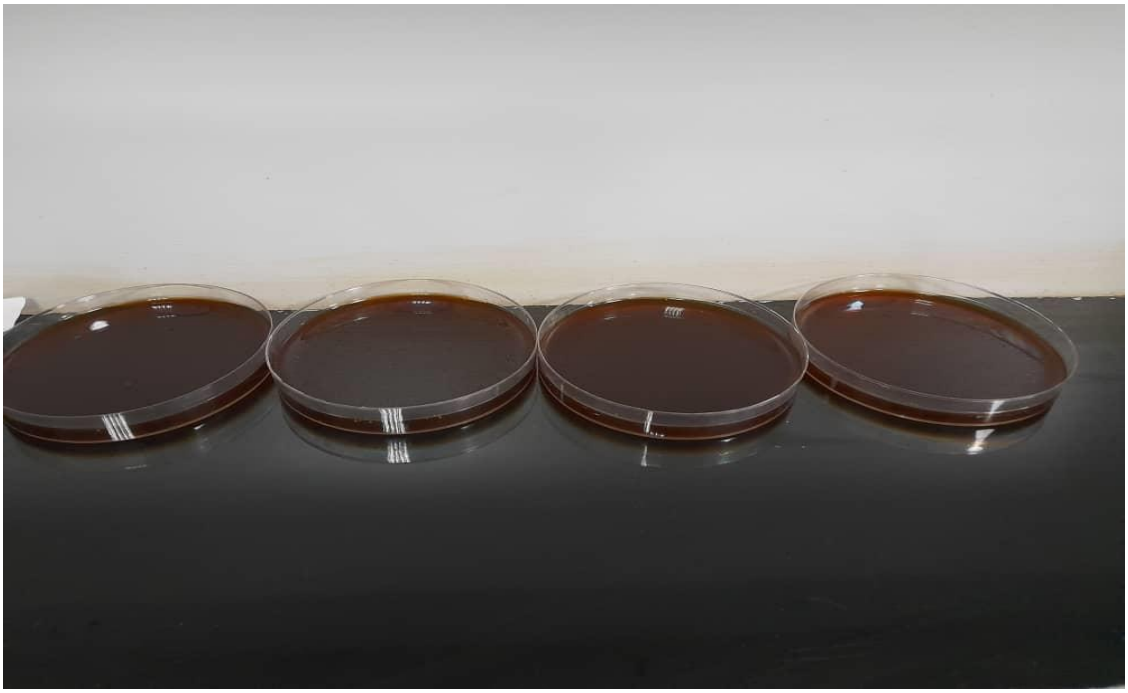
41. World Health Organization (WHO) Expert Committee. Global report on diabetes. 20 Avenue Appia, 1211 Geneva 27, Switzerland: WHO Press, World Health Organization; 2016. Available from: https://www.who.int/publications/i/item/9789241565257. Accessed April17, 2021.Care. 2020;43(5):1048–1056. doi: 10.2337/dc19-2519 .
42. World Health Organization. Health topics/diabetes. 20 Avenue Appia, 1211 Geneva 27, Switzerland:WHO Press, World Health Organization; 2021. Available from: https://www.who.int/health-topics/diabetes#tab=tab_1. Accessed April17,2021.
43. Zengin, G.; Llorent-Martínez, E.; Fernández de Cordova, M.L.; Bahadori, M.B.; Mocan, A.; Locatelli, M.; Aktumsek, A. Chemical composition and biological activities of extracts from three Salvia species: S. blepharochlaena, S. euphratica var. leiocalycina, and S. verticillata subsp. amasiaca. Ind. Crops Prod. 2018, 111, 11–21.

APPENDIXES

Salvia officinalis leaves

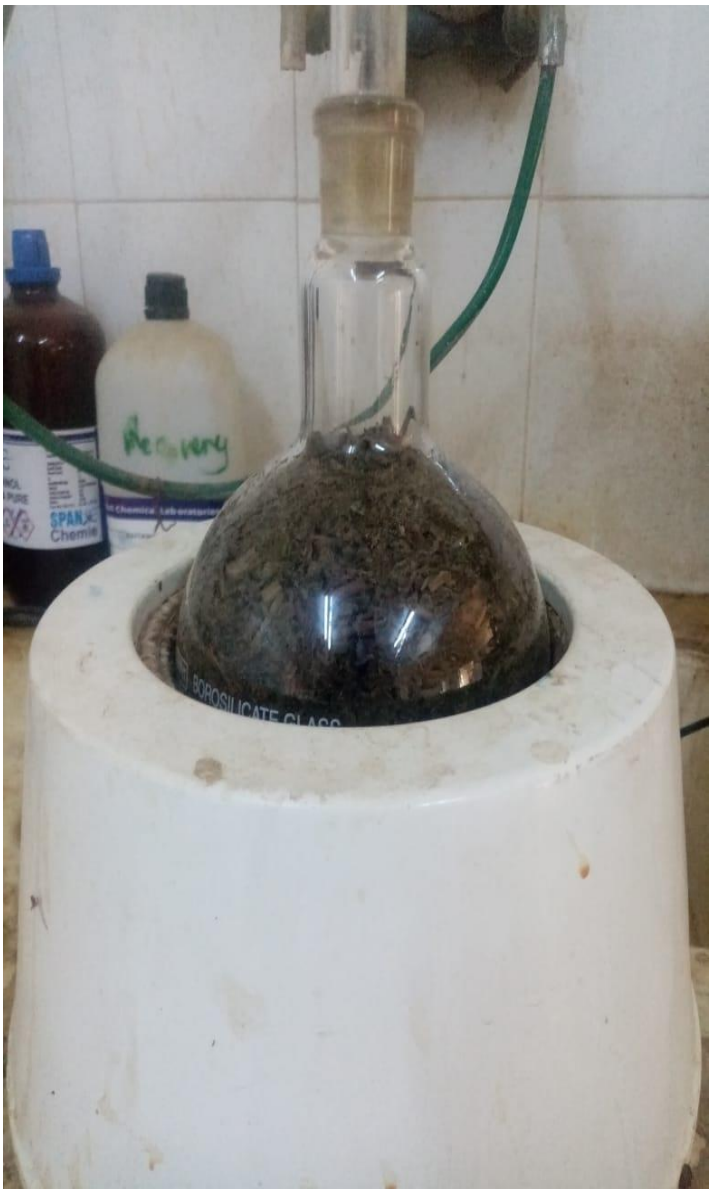
MethanolExtraction

Aqueous extraction



Extraction method

Extraction method



Extraction method

Glucose and glibenclamide

Glucometer and glucometer strip

Albino rat