International Journal of PharmTech Research
CODEN (USA): IJPRIF, ISSN: 0974-4304, ISSN(Online): 2455-9563
Vol.9, No.9, pp 252-260, 2016

Hypoglycemic Effect of *Salvia officinalis* L. Extracts on Induced Diabetic Rabbits

Ahad Kamel Rahiand Lamia, A. M. Al-Mashhady*

Department of Chemistry, College of Science, University of Babylon,Hilla-Iraq

Abstract : This study investigating the hypoglycemic effect of *Salvia officinalis* L. extracts on diabetic Rabbits induced by alloxan and estimated of some biochemical parameters related to oxidative stress. Three groups of six rabbits were used in this study. Group 1 received normal saline orally and considered as a control group, Group 2 was a diabetic group which received a single dose of alloxan 100mg/kg b.w (i.p) and Group3 was a treatment group, received alloxan(100mg/kg b.w.) then treated with 500mg/kg b.w orally of hot extract for four weeks every day. Some parameters such as Glucose-6-phosphate dehydrogenase, malondialdehyde (MDA) and lipid peroxidation were measured as well as the blood glucose levels. *Salvia officinalis* L. hot extract has decreased blood glucose levels and increased the activity of glucose-6-phosphate dehydrogenase in the group 3 compared to the group 2. While the study improved that MDA and lipid peroxidation levels were lower in the treatment group compared with diabetic group.
Keywords: *Salvia officinalis*, Alloxan, Diabetes Mellitus, Glucose-6-phosphate dehydrogenase, Lipid Peroxidation, Malondialdehyde.

Introduction:

Diabetes is a metabolic disorder in which the pancreas in the human body fails to produce insulin, or is unable to use the insulin produced in an effective manner.[1]Diabetes mellitus can be classified in different ways but oneform of classification is as follows (American Diabetes Association, 2004):type I diabetes (Insulin dependent) is due to immune mediated beta-cells destruction, leading to insulin deficiency, Idiopathic diabetes is the type 1 diabetes with no known etiologies and is strongly inherited,type II diabetes (Non-Insulin dependent) is due to an insulin secretory defect and insulin resistance and Gestational diabetes mellitus is any form of intolerance to glucose with onset or first recognition of pregnancy[2].

Diabetesmay be induced by some drugs such as Streptozotocine and Alloxan. Alloxan {(2,4,5,6)tetraoxyhexa hydro pyrimidine} is one of the widely used models to induce diabetes mellitus within the experimental animals. It has been found to be selectively toxic to duct gland beta cells because it preferentially accumulates in the beta cells as glucose analogues. Additionally, the cytotoxic action of Alloxan is mediate mainly by the generation of reactive oxygen species (ROS)[3].

Salvia is the largest genus of plants in the Lamiaceae family. The name *Salvia officinalis* derives from the Latin ‘salveo’, which means “to be saved”. *Salvia officinalis L.* (Lamiaceae, common sage) is a medicalaromatic plant that grows in Portugal as well known for its medical properties. Sage enjoys the reputation of being a panacea[4]. It's having the ability to protect the body against oxidative stress, free radical damages, angiogenesis, inflammation, bacterial and virus infection[5]. Sage tea was effective in the improvement of lipid profile, antioxidant defenses and lymphocyte[6]It has been proposed as effective against cardiovascular

diseases, brain and nervous disorders, various infections anddigestion problems[7].

Glucose-6-phosphate dehydrogenase (G-6-PD), an enzyme expressed in most human tissues is important in the generation of reduced glutathione - a key product in the control of oxidative stress. A low activity of this enzyme in red blood cells leads to Glucose-6-phosphate dehydrogenase deficiency (G-6-PDD). This disease has been overlooked as one of the causes of increased oxidative stress a risk factor for diabetes mellitus a disease which is a threat to the health of many populations[8]. While oxidative stress is a condition in which the cellular production of reactive oxygen species (ROS) exceeds the physiological capacity of the antioxidant defense system. Hyperglycemia is one of the most important factors that are responsible for oxidativestress and the production of ROS in diabetes [9]. MDA is important in the late complications of diabetes mellitus because it contributes to the stiffening of various tissues like cardiovascular tissue[10].

Materials and methods:

Collection of Plant Samples:

The plant materials were brought from local markets in Hilla, the leaves of plant were washed in tap water and dried, then used to create the aqueous and ethanolic extracts that analyzed.

Preparation of the Extracts:

The ethanolic extract, (cold and hot) aqueous extracts were made according to [11].

Phytochemical Constituents:

Chemical tests were organized on the ethanolic extract, aqueous extracts of the plant sample using standard methods. The investigation of phytochemical constituents involved:

1. **Qualitative Study:**It is based on the change the color of the samples[12, 13].
2. **Quantitative Study:** It is based on the precipitationof the sample using standard methods [14].

Determination of the Active Time and Active Dose:

To determine the active dose and active time for the best phytochemical analysis results of extract(ethanolic and hot extracts) of *Salvia Officinals* L. required 12 rabbitsthat be divided the animals as shown in the Table (1)

Table (1):The Categories of Experimental Animals According to Active Dose and Time

Type of Extract	No. of Rabbits	The Dose mg /kg
Hot Extract	3	100
	3	200
	3	500
EthanolicExtract	3	100
	3	200
	3	500
Control	3	Normal Saline

Experimental Design to Study the Hypoglycemic Effect:

Eighteen rabbits of body weight between 500 and 1900 kg were used for the test.The 18 rabbits were fed exclusively on fodder and water to drink and they received no other medication at the time outside of the extract. They were randomly divided into three groups of 6 rabbits treated as follows:

- Group 1 the control group.
- Group 2 considered as a diabetic group and received 100 mg/kg b.w of alloxan (i.p) as a single dose.

- Group3 (the treatment group) received alloxan100mg/kg b.w. Then, treated with 500mg/kg b.w of hot extract for 4 weeks orally every day.

Determination of Biochemical Parameters

To assay blood glucose, glucose-6-phosphate dehydrogenase, MDA and lipid peroxidation in rabbits, we collected blood of these animals from the marginal ear vein, and the blood is collected in dry test tubes and anticoagulant (EDTA for glucose) and then centrifuged at 3000 rpm for 5 minutes and the serum were decanted and stored for the determination of these parameters.

Determination of Blood GlucoseConcentration(mg/dl)

The blood glucose concentration was measured by the enzymatic method with GOD-PAP reagent the blood was determined after four weak of administration of hot aqueous extract *Salvia Officinallis* at zero time and four hours[15].

The principle of thedetermination of glucose is based on the reaction

$$\text{Glucose} + \tfrac{1}{2}\,O_2 + 2H_2O \longrightarrow H_2O_2 + \text{Gluconic acid}$$

Determination of Glucose-6-phosphate dehydrogenase Activity(1U/L):

The principle of this method is based on Beutler method is as follows:

$$\text{G-6-P} + NADP^+ \xrightarrow{\text{G-6-PDH}} \text{6-PGluconate} + NADPH + H^+$$

The rate of NADPH concentration increasing in measured at 340nm is proportional to the G-6PDH activity in the specimen[16].

Determination of MalondialdehydeMDA(µmoles/L) levels

The concept of this procedure was based on spectrophotometric measurement of the color happen through the reaction between thiobarbituric acid and malondialdehyde (MDA) yielding pinkish red chromogen with an absorbance at 532 nm[17].

Determination of Lipid PeroxidationAssay(µmole/L)

A modified thiobarbituric acid – reactive kinds assay was used to evaluate the lipid peroxide formed using egg-yolk homogenates as lipid – rich media[18].

Statistical Analysis:

Statistical analysis was performed bySPSS statistics version 17. Subject with groups (1, 2 and 3) were compared between them. Means, standard deviation, confidence interval 95% the results of biochemical analysis were assessed by student's t-test; significant variation was considered when the P value was less than 0.05.

Phytochemical Analysis

After preparation of different extracts the results show that the yield of ethanolic extract was (8.3%).While the yield of the hot and cold extracts are (3.83 %w/w) and (3.80 %w/w) respectively. These results are due to the effect of heating and the nature of the solvent which may be isolated most of the active chemical components in ethanolic extract more than aqueous cold and hot extract.

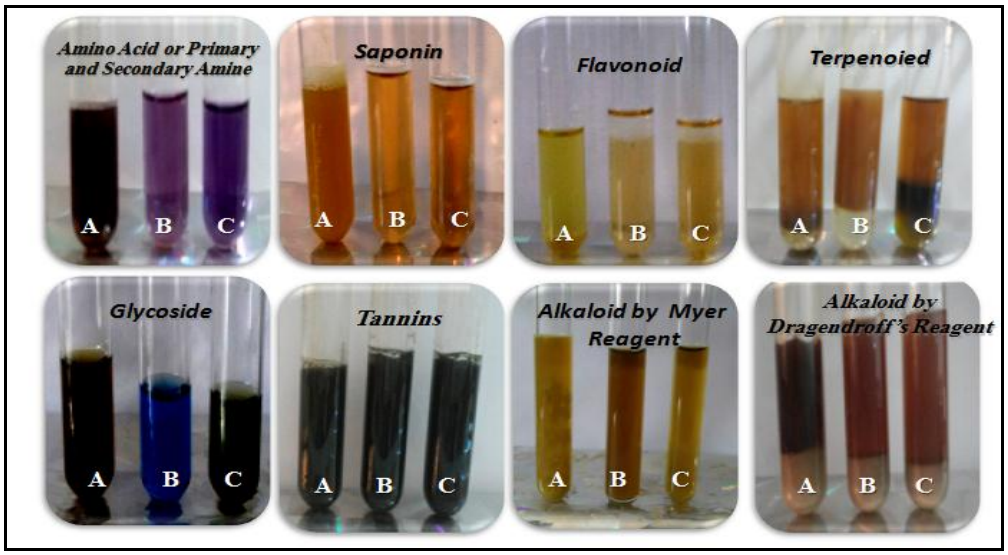
Phytochemical analysis is very helpful in the estimation of some active biological components of medicinal plants [19,20]. *Salvia officinalis* L. extracts showed positive results for all constituents analyzed, except for anthroquinones, phlobatanins as shown in Table (2) and Figure (1).

Medicinal plants have bioactive compounds which play a great role in healing and are used for curing of various human diseases[21]. Phytochemicals have a main role in inhibiting chronic diseases like cancer, diabetes and coronary heart disease. The major classes of phytochemicals with disease inhibition functions are dietary fiber, antioxidants, anticancer, detoxifying agents, inviolability potentiating factor and neuropharmacological agents[22].Kumari S J *etal*. introduced a review to elucidate that natural compounds like terpenoids, alkaloids, phenolic compounds such as flavanoid have shown antidiabetic effect through the insulin like activity. Among the reviewed compounds, flavonoids majorly exhibit the Antidiabetic activity. It acts by preventing β-cell apoptosis and promotes β-cell proliferation and insulin secretion[23].

Table (2): The Qualitative Analysis of *Salvia Officinal* L. Extracts

Phytochemical Constituents	Ethanolic Extract	Hot Extract	Cold Extract	Color
Alkaloids	+++	++	+	Turbidity
Amino Acids or Primary and Secondary Amine	+	++	++	Blue - Violet
Anthroquinones	-	-	-	-
Flavonoids	++	+	+	Yellow
Glycosides	-	+++	-	
Phlobatanins	-	-	-	----
Saponins	+++	++	++	Forth
Tannins	+++	+++	+++	Blue-black
Terpenoids	++	++	++	Reddish Brown

Where as: (+) Present, (++) Fairly Good Amount, (+++) Good Amount, (-): Absent



Figure(1): Qualitative Analysis of Phytochemical Constituents for *Salvia officinal L.* Extracts. Whereas:- A: Ethanolic Extract B: Hot Extract C: Cold Extract

While the results of quantitative analysis of phytochemical screening of *Salvia officinalis* L.extracts showed in the Table (3).

Table (3): The Quantitative Analysis of Phytochemical Screening of *Salvia Officinalis* L. Extracts

Cold Extract	Hot Extract	Ethanolic Extract	Phytochemical Constituents
0.56%	**0.56%**	**0.77%**	**Alkaloids**
1.3%	**1.6%**	**1.4 %**	**Flavonoids**
2.82%	**3.79%**	**2. 92%**	**Saponins**

Determinationof the Active Time and Active Dose

The study included the determination of active dose and time for the hypoglycemic effect of *Salvia officinalis* L. extracts (hot and ethanolic 70%) from the qualitative and quantitative analysis of *Salvia officinalis* L. extracts which illustrated that hot and ethanolic extract given the best results of phytochemical constituents in the Table (4) show the hypoglycemic effect of the two extracts.

Table (4): Hypoglycemic Effect of *Salvia Officinalis* L.in Different Times (0, 2, 4, 6 and 24) Hours(The results were expressedas mean of data for three rabbits in each group).

Control	Blood Glucose Levels for Doses (mg/dl)				Extracts
N.S	500mg/kg	200 mg/kg	100 mg/kg		
103	120.6	103	92.3	0	Hot Extract
97	74.8	74	81	2	
94	69.6	70	73	4	
99	108	103.6	92.2	6	
111	103	101.2	97	24	
103	122.6	93	105	0	Ethanolic Extract
97	106	78.8	96.6	2	
94	97	77	74.6	4	
99	99	126	116.6	6	
111	102	124	121	24	

The blood was collected from the rabbits of the groups, at different times 0,2,4,6,24 hours. The resultsin the Table (4) showed that four hours is the best time for hypoglycemic effect for the two extracts (hot and ethanolic 70%) after six hour the blood glucose of rabbit was significant increased compared with four hours of oral administration the two extracts give good results for hypoglycemic effect but ethanolic extract cause several deaths cases for the rabbits especially in 200 mg/kg and 500 mg/kg while the treatment with hot extract didn't cause any death for that, the hot extract will be chosen to study the hypoglycemic effect of *Salvia Officinalis* L. on induced diabetic rabbits. Also Table (4) showed that the glucose levels are directly proportional with concentration of hot extract.

Estimation of Blood Glucose Concentration (mg/dL)

Diabetes in experimental animal occurs due to a high oxidative stress resulting from the persistent and chronic hyperglycemia[24].

Table: (5) Glucose Levels (mg/dl) for Rabbits Groups Treated with Hot Aqueous Extract of *Salvia Officinallis* Extract Compared with Control Group

P-Value	Confidence interval		S.E	Mean ± S.D	The Groups
	Upper	Lower			
At Zero Time					
0.003*	104	80	3.65	91.3±8.9	Group 1
0.005**	312	284	4.75	292±11.5	Group 2
0.007***	160	140	3.34	156±9.3	Group 3
After Four Hours					
0.001*	96	80	2.46	86.6 ± 6.0	Group 1
0.001**	320	288	4.56	303.3 ± 11.1	Group 2
0.002***	120	104	3	115.3 ± 7.3	Group 3

: it means significance related of G1with G2.**: it means significant related of G1 with G3 it means significant related of G2 with G3.**

The results showed in the Table (5) that refer to the ability of the aqueous extract of *Salvia Officinal's* in reducing of glucose levelsand this results give a good indicator for the hypoglycemic effect of sage that related to the active component of the plant,which have been found to stimulate secretion or possess an insulin like effect [25], and can act on pancreatic β-cells leading to their proliferation and secretion of more insulin, and these flavonoids may exert its effects on insulin release from islet of langerhans via changes in Ca^{2+} metabolism. The active component in the aqueous extract of *Salvia Officinal's* may be responsible for β-cell regeneration and insulin release. Also, will promote induction of hepatic glucokinase, and the increases in this enzyme activity cause directly demonstrates increased insulin release from β-cell[26].This resultagrees with other previous studies[27].

Determination of Blood glucose-6-phosphate dehydrogenase Activity:

Diabetes mellitus is a common and complicated disease. Studies imply blood glucose and its **oxidize** derivatives have a key role in the pathologic process of DM. "Glucose-6-phosphate dehydrogenase" (G6PD), was antioxidant enzyme and an important in preventing its complications. Unsuitable management of blood glucose decreases G6PD activity and willincrease diabetes mellitus complications[28].

High glucose has been shown to extendROS in several cell types with diabetes because of a combination of increased production of ROS along with decreased antioxidant function [29]. Many laboratories have shown that pancreatic cells are very sensitive to oxidant damage, which has been attributed to the low expression levels of antioxidant enzymes. Thus, cells are likely at higher risk of oxidizer mediated cellular injury and death as compared to alternative class cell varieties[30]. Table (6) illustrated the G6PD for the rabbits groups

Table(6): G-6-PDH Activity (IU/L) for Rabbits Groups Treated with Hot Aqueous Extract of *Salvia Officinallis*Compared with Control Group

P-Value	ConfidenceInterval		S.E	Mean ± S.D	The Groups
	Upper	Lower			
At Zero Time					
0.0005*	4.92	3.93	0.125	4.505±0.37	Group 1
0.0007**					
0.087***	3.44	2.95	0.08	3.03±0.2	Group 2
0.087****	3.44	2.95	0.08	3.19±0.26	Group 3
After Four Hours					
	3.44	2.95	0.15	3.35 ± 0.36	Group 3

: it means significance related of G1with G2,**: it means significance related of G1with G3 at zero time, ***: it means significance related of G2with G3 at zero time, *: it means significance related of G3at zero time with G3 at four hours.**

The results showed in the above Table explain that the diabetic group has the lowest value, from Glucose-6-phosphate dehydrogenase (G6PD) activity is the main source of the major intracellular reluctant, NADPH, which is required by many enzymes, including enzymes of the antioxidant pathway, high glucose impairs G6PD activity in endothelial and kidney cells,which leads to decreased cell survival. Pancreatic cells are highly sensitive to increased ROS[27] .

Determination of Malondialdehyde MDA (µmoles/L) levels

Malondialdehyde (MDA) is an accepted marker of lipid oxidative damage. Malondialdehye was produced when highly reactive oxygen metabolites, particularly hydroxyl radicals, act on unsaturated fatty acidsof phospholipids components of membranes[9].

Table (7): Malondialdehyde Levels (µmoles/L) for Rabbits Groups Treated with Hot Aqueous Extract of *Salvia Officinallis* Extract Compared with Control Group

P-Value	Confidence Interval		S.E	Mean ± S.D	The Groups
	Upper	Lower			
At Zero Time					
0.01*	**1.4**	**1.2**	**0.0429**	**1.3±0.1**	**Group 1**
0.046**					
0.05***	**3.5**	**2.6**	**0.127**	**3.06±0.30**	**Group 2**
0.001****	**2.7**	**1.8**	**0.125**	**2.36±0.3**	**Group 3**
After Four Hours					
	1.9	**1.2**	**0.095**	**1.45±0.23**	**Group 3**

: it means significance related of G1with G2,**: it means significance related of G1with G3at zero time, ***: it means significance related of G2with G3at zero time , * : it means significance related of G3at zero time with G3at four hours**

In the Table (7) we found a significant rise in serum level of MDA in the group ofdiabetes mellitus as compared to controls, indicating that an increase in oxidative stress might play a key role in pathogenesis of diabetes mellitus and its complication.

Determination ofLipid Peroxidation Assay(µmole/L):

Lipid peroxidation is employed as a marker of cellular oxidative stress and contributes to the oxidative damage that occurs as a result of xenobiotics metabolism, inflammatory processes, ischemia, reperfusion injuries and chronic diseases such as atherosclerosis and cancer[31]. There's considerable evidence that hyperglycemia represents the mostexplanation for complications of diabetes (DM), and oxidative stress ensuingfrom an increased generation of reactive oxygen species plays a vital role in their pathologic process[32].

Table (8): Lipid Peroxidation Levels (µmoles/L) for Rabbits Groups Treated with Hot Aqueous Extract of *Salvia Officinalis*L. Extract Compared with Control Group

P-Value	Confidence Interval		S.E	Mean ± S.D	The Groups
	Upper	Lower			
At Zero Time					
0.0007*	**29**	**23**	**0.98**	**24.8±2.4**	**Group 1**
0.00009**					
0.297***	**39**	**31**	**1.18**	**35.1±2.9**	**Group 2**
0.06****	**39**	**30**	**0.8**	**33.8±3.5**	**Group 3**
At Four Hours					
	34	**28**	**0.8**	**31.6±1.9**	**Group 3**

It means significance related of G1with G2,: it means significance related of G1with G3at zero time, ,***: it means significance related of G2with G3at zero time , ****: : it means significance related of G3at zero time with G3at four hours**

At the Table (8) we found a significant rise in serum level of lipid peroxidation in the group of diabetes mellitus as compared to controls.

References:

1. Abhinov T., MdAasif A.K., Ashrafa ., Parveen S., and Kumar K.P.(2013). Diabetes epidemic in India: risk factors, symptoms and treatment.Indian Journal of Research in Pharmacy and Biotechnology. 1(2):234-243.
2. American Diabetic Association. Diagnosis and classification of Diabetes mellitus. Diabetes care, 33(1), 2010, S62-S65.
3. Rohilla A. and Ali S.(2012). Alloxan Induced Diabetes: Mechanisms and Effects. International Journal of Research in Pharmaceutical and Biomedical Sciences;3 (2):819-822.
4. Abdallah I. Z. A., Khattab A.H., Sawiress F. A.R., andEL-Banna R.A.S.(2010). Effect of salvia officinalis L. on osteoporotic changes in aged Non-cycling female rats. Med.J.Cario.univ;78(1):1-9.
5. Hamidpour R., Hamidpour S., Hamidpour M. and Shahlari M. (2013). 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. Global Journal of Medical research; 13(7):1-8.
6. Sá C.M , Ramos A.A. , Azevedo M.F , Lima C.F. , Fernandes-Ferreira M. and Pereira-Wilson C. (2009). Sage Tea Drinking Improves Lipid Profile and Antioxidant Defences in Humans. International Journal of Molecular Sciences; *10*: 3937-3950.
7. Khattab H. A. H., Mohamed R.A. and Hashemi J.M.(2012). Evaluation of Hypoglycemic Activity of *Salvia officinalis L.* (Sage) Infusion on Streptozotocin-Induced Diabetic Rats . Journal of American Science;8(11):411-416.
8. Adinortey M. B.,.Owusu R. K., Galyuon I. K. A., Ekloh W., Owusu I. and Larbi D. A. (2011). G-6-PD deficiency - a potential risk factor for development of diabetes mellitus Journal of Medicine and Medical Science; 2(8) 1017-1021 .
9. Deokar P., Jagtap A., and Yerawar C.(2016) Correlation of protein carbonyl and MDA in diabetes and its complications. Indian Journal of Basic and Applied Medical Research; 5(2):284-289.
10. Slatter D.A., Bolton C.H. and Bailey A. J. The importance of lipid-derived malondialdehyde in diabetes mellitus. Diabetologia Springer-Verlag ;43:550-557.
11. Sharma V. K., Kumar S., Patel H. J. and Hugar S. (2010). "Hypoglycemic Activity of Ficus*Glomerata* in Aloxan Induced Diabetic Rats". International Journal of Pharmaceutical Science Review and Research; 1(2):18-22.
12. Majaw S. and Moirangthem J. (2009). "Qualitative and Quantitative Analysis of Clerodendroncolebrookianum Walp. Leaves and ZingibercassumunarRoxb. Rhizomes". Ethnobotanical Leaflets; 13:578-589.
13. Hawk P. B., Oser B. L. and Sumerson H. W. (1954). "Practical physiological Chemistry". 13th edition. Mcgraw-Hill, Book Company, INC. New York: 63-172.
14. Aliyu A. B., Musa A. M., Oshanimi J. A., Ibrahim H. A. and Oyewale A. O. (2008). "Phytochemical Analyses and Mineral Elements Composition of some Medicinal Plants of Northern Nigeria". Nig. Journ. Pharm. Sci.; 7(1): 119– 125.
15. Tietz N.W.(1995) Clinical Guide to the laboratory tests 3rded W.B Philadephia saunders.422-447.
16. Beutler E.(1984).Red cell metabolism: ameasure of biochemical methods 3rdedOriando .Grune et Stratton 68-70.
17. Guidet B. and Shah S.v.(1989).Enhanced in vivo H_2O_2 generation by rat kidney in glycerol – induced renal failure. American Journal of Physiology; 1257,: 440 – 444.
18. Pandey N. Chaurasia J. K. Tiwari O. P. and Tripathi Y. B. (2007). Antioxidant properties of different fractions of tubers from Puerariatuberosa Linn. *Food Chemistry*; 105: 219–222.
19. Al-Temimi S. M. and Al-Mashhedy L. A. M.(2015) Estimation of the Phytochemical Constituents and Biological Activity of Iraqi Ocimum sanctum L Extracts. International Journal of Pharma and Bio Sciences; 6 (1): 999-1007.
20. Al-Kawaz H. S. and AL-Mashhady L. A. M. (2016). Evaluation of the Phytochemical Constituents and Oxidant – Antioxidant Status for Actinidiadeliciosa Extracts. International Journal of Pharmacy & Therapeutics; 7(1): 31-41.
21. Wadood A., Ghufran M., Jamal S.B. , Muhammad Naeem M. , Khan A., Ghaffar R., and Asnad.(2013). Phytochemical Analysis of Medicinal Plants Occurring in Local Area of Mardan. Biochemistry & Analytical Biochemistry;2 (4):2-4.
22. Saxena M. Saxena J. Nema R. Singh D. and Gupta A.(2013). Phytochemistry of Medicinal Plants.

Journal of Pharmacognosy and Phytochemistry;1(6): 168-182.
23. Kumari S J., Sangeetha M, Pavithra R.(2016).A Retrospective Review on Indian Traditional Herbs and its Biocompounds in Diabetes.;International Journal of PharmTech Research,9(5): 444-460.
24. Pitocco D., Tesauro M., Alessandro R ., Ghirlanda G ., and Cardillo C.(2013). Oxidative Stress in Diabetes: Implications for Vascular and Other Complications. International Journal of Molecular Sciences.14:21525-21550.
25. Eidi M[1], Eidi A, Zamanizadeh H.(2005). Effect of Salvia officinalis L. leaves on serum glucose and insulin in healthy and streptozotocin-induced diabetic rats.J Ethnopharmacol. 100(3):310-3.
26. Hasaneina P, Felehgari Z, Emamjomeh A.(2016). Preventive effects of Salvia officinalis L. against learning and memory deficit induced by diabetes in rats: Possible hypoglycaemic and antioxidant mechanisms.Neuroscience Letters 622 : 72–77.
27. GuptaSh, and Mukherjee M..(2014).Diabetes mellitus and its treatment with some traditional herbs from the different districts of West Bengal: A Review.; International Journal of PharmTech Research; 6(6):1941-1949.
28. El-Abhar H. S. and Schaalan M. F. (2014). Phytotherapy in diabetes: Review on potential mechanistic perspectives World J Diabetes. 5(2): 176–197.
29. Zhang Z., Liew C. W., Handy D. E., Zhang Y., Leopold J . A., Hu J., Guo L. , Kulkarni R. N., Loscalzo J., and Stanton R .C.(2010). High glucose inhibits glucose-6-phosphate dehydrogenase, leading to increased oxidative stress and -cell apoptosis. The FASEB Journal 24:1497-1505.
30. Lupi, R., and Del Prato, S. (2008) Beta-cell apoptosis in type 2 diabetes: quantitative and functional consequences. Diabetes Metab. 34, S56–64.
31. KaurinovicB.andPopovic M.(2012). Biochemistry, genetics and Molecular Biology.Lipid peroxidation, book edited by Angel CatalaISBN 978-953-51-0716-3, Published: August 29, 2012 under CC BY 3.0 license.Liposomes as a Tool to Study Lipid Peroxidation. intech open science.P: 155-156.
32. Davì G. Falcoa., and Patrono C. (2005)Lipid Peroxidation in Diabetes Mellitus. Antioxidants & Redox Signaling, 7(1):256-258.

*****