Phytochemical Screening, Acute Toxicity and Antidiabetic Activity of Ethanolic Extract of *Salvia officinalis* L. in Wistar Rat

Rachida BOUTELDJA[1,2] (✉)
Radhouane DOUCENE[3]
Hebib AGGAD[1]
Fatima Zohra ABDI[4]
Hamza BELKHODJA[4]
Aicha BELAL[5]
Mustapha ABDALI[1]
Khaled ZIDANE[3]

Summary

Salvia officinalis L. is an aromatic herb that is widespread in the Tiaret region of Algeria where it plays a major role in the translational pharmacopoeia. This work aims at phytochemical screening, studying of acute toxicity and evaluation of antidiabetic activity of ethanolic extract of *S. officinalis* in Wistar rat induced by a diabetogen "Alloxan". First, the phytochemical characterization was carried out by determination of the main bioactive compounds. It was followed by studying of acute toxicity in Wistar rat. Then, the evaluation of antidiabetic activity was performed by induction of diabetes in Wistar rat with Alloxan. The results showed the richness of the ethanol extract of *S. officinalis* in different bioactive compounds such as tannins, flavonoids, cardiac glycosides and alkaloids. The absence of serious clinical signs or dead rats during the observation period indicates that the ethanol extract of *S. officinalis* administered *per os* is devoid of acute toxicity in rats at 2000 mg kg^{-1} body weight. For the antidiabetic activity, results reported a highly significant reduction in blood glucose, cholesterol levels at a dose of 300 mg kg^{-1} in diabetic rats.

Key words

Salvia officinalis L., diabetes, alloxan, polyphenols, toxicity, antidiabetic activity

[1] Laboratory of Hygiene and Animal Pathology, Institute of Veterinary Sciences, University of Tiaret, Algeria

[2] Faculty of Natural and Life Sciences. Ibn Khaldoun University of Tiaret, Algeria

[3] Laboratory Reproduction of Farm Animals, Institute of Veterinary Sciences, University of Tiaret, Algeria

[4] Laboratory of Bioconversion, Microbiology Engineering and Health Safety, University of Mustapha Stambouli, Mascara, Algeria

[5] Laboratory of Medical Analysis, Tiaret, Algeria

✉ Corresponding author: bouteldjarachida.92@gmail.com

Received: June 3, 2022 | Accepted: July 13, 2023

Introduction

Diabetes mellitus is a real health problem that affects all categories of humans characterized by chronic hyperglycemia accompanied by disturbances in carbohydrate, lipid and protein metabolism resulting from impaired insulin function through absence or deficiency (Kahn, 2005). This pathology is linked to several agents including environmental factors, smoking, alcohol consumption, lack of physical activity, exposure to enteroviruses and immune cell damage (Frank et al., 2001; Raman, 2016). Thus, it may be related to oxidative stress causing molecular damage from the oxidation of lipids, proteins and nucleic acids (Favier, 2006).

Several chronic complications cause this disease, including damage to small blood vessels including retinopathy or eye disease, kidney disease called nephropathy and neural lesions or neuropathy. Chronic complications of diabetes also include depression, dementia and sexual dysfunction (Forbes et Cooper, 2013). To reduce these complications and minimize the side effects of conventional drugs, researchers are moving towards the use of more reliable natural sources through the application of medicinal plants thanks to their wealth of bioactive compounds. Several studies prove the chemo-preventive effect of these products against several pathologies such as cancer, diabetes, obesity and Alzheimer's disease. (Ben Younes, 2018; Hartman, 2018).

Sage (*Salvia officinalis* L.) is a shrubby perennial herb native to the Mediterranean and Balkan regions. Their extracts are used in the medical field thanks to their antioxidant, antibacterial, anti-inflammatory, anti-cancer and anti-diabetic effects and because they are rich in bioactive compounds (Ghorbani et Esmaeilizadeh, 2017). This work aims to study the polyphenolic extract of *S. officinalis* by phytochemical screening, determination of their toxicity and antidiabetic activity in vivo in an animal model induced with a chemical diabetogen.

Materials and Methods

Plant Material

Leaves and flowers of *S. officinalis* was collected in April in the region of Tiaret (Algeria) (35° 23' 17" North, 1° 19' 22" East). It was identified at the botanical laboratory of the National Superior School of Agronomy (Algiers). It was washed and then dried in the open air, crushed with an electric grinder and stored at room temperature, away from humidity and light.

Animals of Experimentation

24 adult male Wistar rats with a weight of 246.87 ± 22.88 g were provided from the Pasteur Institute of Algiers. The rats were placed for 15 days at the laboratory of Animal Hygiene and Pathology at the Institute of Veterinary Sciences "Tiaret-Alger", under well controlled conditions: temperature of 20 ± 2 °C and a light/dark cycle 12h/12h under a standard commercial diet. The procedures used during these studies were in accordance with the European Directive Concerning Animal Testing (Directive 2010/63/EU) (decision N°: L276/33).

Preparation of Ethanolic Extract

The ethanolic extract was prepared by maceration with a ratio of 5 g of powder mixed with 50 ml of ethanol (80%) under agitation for 24 hours at room temperature. Then, the mixture was filtered on filter paper and the filtrate evaporated and dried at 40 °C (Ghezelbash et al., 2015).

Phytochemical Screening

The phytochemical characterization of ethanolic extract of *S. officinalis* was carried out by different classical methods as shown in table 1.

Acute Toxicity Test

The acute toxicity test of ethanolic extract of *S. officinalis* was performed according to the Organization for Economic Co-operation and Development (OECD). A single dose of 2000 mg kg^{-1} body weight of ethanolic extract of *S. officinalis* was administered *per-os*. A total of 8 rats were divided into two groups of four rats each: Group 1: control group that received distilled water (CG^-). Group 2: received ethanolic extract of *S. officinalis* (GTES). After the administration of extract, rats were observed for any changes in behavior, color of the fur, mucous membranes and eyes after 30 min, 1, 2, 3 and 24 hours and then daily observed up to 14 days (Raju et Reddy, 2017). After 14 days, the rats were sacrificed and blood samples were taken for biochemical analyses: blood glucose, cholesterol, triglycerides, glutamate-oxaloacetate-transaminase (GOT), glutamate-pyruvate-transaminase (GPT), urea and creatinine. Organs (kidneys, heart, lung, small intestine and colon) were taken for macroscopic examination and lesions determination.

Antidiabetic Activity

Diabetes was inducted by the administration of a single dose of a diabetogenic chemical solution (Alloxan) at a dose of 120 mg kg^{-1} body weight intraperitoneally. After 72 hours, a fasting blood glucose test was performed using a Glucometer (BIONIME GM 550). Rats with blood glucose levels above 200 mg dL^{-1} were selected for experimentation. 16 rats were divided into four groups, four rats each. Group 01: Control group (non-diabetic rats) received distilled water (CG^-); Group 02: Diabetic rats without treatment (GDW); Group 03: Diabetic rats treated with glibenclamide (10 mg kg^{-1} body weight) (GDGL) and Group 04: Diabetic rats treated with ethanolic extract of *S. officinalis* at a dose of 300 mg kg^{-1} body weight (GDES). Body weight, food intake and blood glucose levels were measured weekly. These treatments were applied during a month, when the blood glucose test was performed. 15 days later, the rats were sacrificed and the blood tests were achieved (glycemia, urea, creatinine, cholesterol, triglycerides, glutamate-oxaloacetate-transaminase (GOT) and glutamate-pyruvate-transaminase (GPT)).

Statistical Analysis

The results were presented as an average ± SEM. The statistical processing of the data was carried out using STATISTICA software (version 8.0.725.0). $P < 0.05$ was used as the significance level.

Table 1. Phytochemical analyses of ethanolic extract of *S. officinalis* L.

Bioactive compounds	Methods	Results
Alcaloids	Bouchardat test : 2 mL of an extract solution + 2 drops of BOUCHARDAT reagent (Soni et Sosa, 2013). Wagner test : 2 mL Extract + 2 mL Wagner reagent (Bagre et al., 2007).	A reddish-brown precipitation
Tanins	2 ml of extract +1 to 2 drops of ferric chloride solution ($FeCl_3$) diluted at 0,1% (Harborne, 1998).	A dark green color for catechic tannins and blue-green for gallic tannins
Saponins	10 ml of extract in a test tube. The tube is shaken for 15s and then left to stand for 15 min (Bidie et al., 2011)	A height of persistent moss, greater than 1cm
Terpenoids	salkowski test: 0.2 g of extract plus 2 mL of chloroform + 3 mL of concentrated sulfuric acid (Mujeeb et al., 2014)	Formation of a rusty coloration layer at the interface
Cardiac glycosides	2 mL of extract was dissolved with 2 mL of chloroform and concentrated sulfuric acid (Soni et Sosa, 2013)	A dark reddish-brown interfacial layer formation
Anthocyanines	1 mL extract + 3 mL H_2SO_4 (10%) and 1 mL NH_4OH (10%)	A coloration turns to blue
Flavonoids	Extract was treated with a few drops of lead acetate solution (Tiwari et al., 2011)	The formation of yellow precipitate
Mucilages	1 mL of extract is added to 5 ml of absolute ethanol (Noudogbessi et al., 2013)	The appearance of a flaky precipitate

Results and Discussion

Phytochemical Screening

Phytochemical screening results for ethanolic extract of *S. officinalis* are presented in Table 2.

Table 2. Phytochemical screening results of ethanolic extract of *S. officinalis* L.

Bioactive compounds	Results
Tanins	+
Flavonoids	+
Terpenoids	-
Mucilages	-
Anthocyanines	-
Alcaloids	+
Cardiac glycosides	+
Saponins	-

Note: +: presence ; - : absence.

The results were in agreement with those obtained by El Ouali et al. (2016). These confirmed the richness of the ethanolic extract of *S. officinalis* in flavonoids, tannins, glycosides, mucilages. Khare et al. (2019) reported the presence of glycosides, alkaloids and flavonoids in the methanolic extract of *S. officinalis*. Other research revealed the richness of the methanolic and aqueous extracts of *S. aegyptiaca* L. in alkaloids, flavonoids, steroids, glycosides, tannins, terpenoids and saponins (Krimat et al., 2015; Pratrima et al., 2017). The work of Kadhim et al. (2016) revealed that the aqueous extract of *S. officinalis* was rich in bioactive compounds such as tannins, alkaloids, flavonoids, cardiac glycosides, steroids and saponins.

Acute Toxicity Test

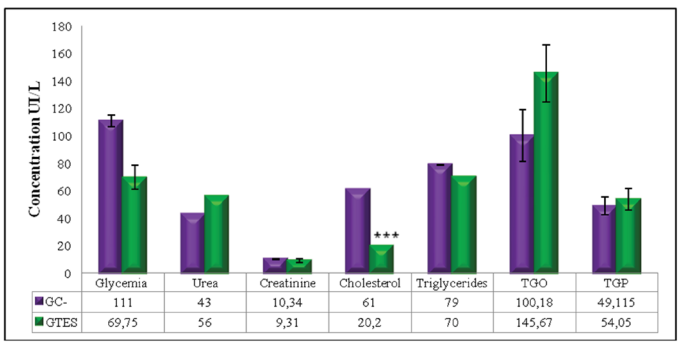
The rats used to evaluate the toxic effect of ethanolic extract of *S. officinalis* had an initial weight of 266.67 ± 14.3 g and were treated for a period of four weeks with a standard diet. At the end of the experiment the groups (CG^-) and (GTES), reached a weight and food intake according to plan , as presented in table 3. On the other hand, blood tests are shown in Fig. 1.

Results showed a highly significant decrease with $P \leq 0.001$ GTES vs GT- for blood glucose and cholesterol (111 ± 8.04 vs 69.75 ± 17.63 mg dL^{-1}) and (61 ± 11 vs 20.25 ± 13.91 mg dL^{-1}). This reduction can be explained by the hypoglycemic and hypocholesterolemic effect of ethanolic extract of *S. officinalis* (Alarcon-Aguilar et al., 2002). On the other hand, no significant differences were observed for urea, creatinine, triglycerides, TGO and TGP (56 ± 13 mg dL^{-1}; 9.31 ± 2.60 mg dL^{-1}; 70 ± 5 mg dL^{-1}; 145.68 ± 41.85 IU/l and 54.05 ± 15.91 IU/l, respectively) and (43 ± 3 mg dL^{-1}; 10.34 ± 0.92 mg dL^{-1}; 79 ± 21 mg dL^{-1}; 100.19 ± 37.86 IU/l and 49.15 ± 13.4 IU/l, respectively) for GTES and GC, respectively.

Table 3. Evaluation of experimental rats mass and food intake during the experiment

	Initial weight (g)	Final weight (g)	Weight Gain (g)	Food Intake (g per week)
CG$^-$	273.6 ± 11.74	271.21 ± 5.1	-2.39	248.51 ± 10.09
GTES	259.74 ± 14.54	273.71 ± 13.03	-13.97	210.88 ± 17.63

Note: CG - control group; received distilled water; GTES - group 2 Received ethanolic extract of *Salvia officinalis* L.



Note: glycemia, urea, cholesterol and triglycerides are expressed in mg dL^{-1}. Creatinine in mg L^{-1} and TGO with TGP in IU/l; *** - a highly significant difference of GTES group vs GC$^-$

Figure 1. The results of the balance of the analyses after the sacrifice (means ± SEM with n = 4)

These results were similar to those obtained in the study of Li et al. (2010) which confirmed the non-toxicity of ethanolic extract of *S. przewalskii* Maxim. The administration of aqueous extract of *S. scutellarioides* at a dose of 2000 mg kg^{-1} did not cause any adverse effects in rats and mice, including behavioral changes, weight variation, loss of appetite, imbalance in blood sugar, creatinine, cholesterol and triglyceride levels and macroscopic organ abnormalities (Ramirez et al., 2007).

Gebru et al. (2014) showed that observation of mice for 24 hours up to 14 days after administration of methanolic extract of the aerial part of *S. tiliifolia* Vahl at a dose of 2000 mg kg^{-1} provided no signs of toxicity including weight loss, lack of appetite and cases of death. In the light of these results, it was concluded that the administration of ethanolic extract of *S. officinalis* at a dose of 2000 mg kg^{-1} body weight did not declare any toxicity (behavioral changes, loss of appetite, agitation, digestive disorders and blood balance) with the total absence of mortalities during the 15 days of experimentation.

Antidiabetic Activity

Evolution of Body Weight

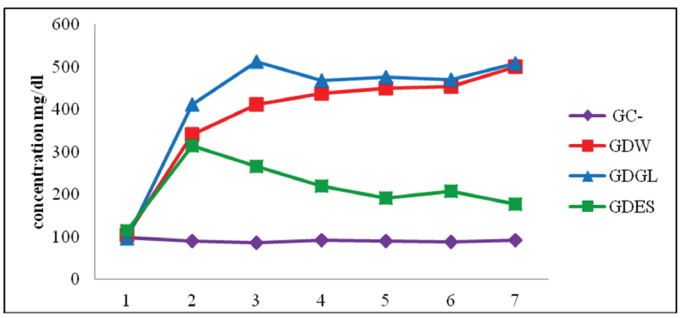
The body weight monitoring showed a highly significant weight reduction ($P \leq 0.001$) in the groups of diabetic rats without treatment (GDW), treated with glibenclamide (GDGL) and the group receiving the ethanolic extract of *S. officinalis* (GDES) compared to the control (GC$^-$). At the same time, a highly significant increase in the amount of food ingested was reported in the group of diabetic rats without treatment (GDW) compared to the group of diabetic rats treated with glibenclamide (GDGL) and the group of rats receiving the ethanolic extract of *S. officinalis* (GDES) and significant with the negative control group (GC) (Table 4).

These results were corroborated with the work of Ewenighi et al. (2018). It was reported that the chemical induction of diabetes caused an increase in food intake with weight loss in diabetic rats. At the same time, a slight increase in weight was observed in rats receiving the ethanolic extract of *S. officinalis*. This was confirmed by Eidi et al. (2011) who exposed the protective effect of ethanolic extract of *S. verticillata* against weight loss in diabetic rats.

Marcelo et al. (2017) explained that the exaggerated dietary ratio due to glucose loss in the urine caused the body to demand more calories from the food ingested, while the decrease in weight may be due to the degradation of structural protein and muscle tissue (Oliveira et al., 2013).

Biochemical Parameters

Results of glycemia levels during experimentation showed a highly significant increase ($P \leq 0.001$) in the groups of diabetic rats without treatment and the group treated with glibenclamide (GDW and GDGL) compared to the Control group (GC$^-$) of (393.85 ± 127 versus 93.93 ± 7.6 mg dL^{-1}) and (420.32 ± 146 versus 93.93 ± 7.6 mg dL^{-1}) respectively. A significant reduction ($P \leq 0.05$) was seen between the group of rats that received ethanolic extract of *S. officinalis* (GDES) of 213.03 ± 64.16 mg dL^{-1} versus (GDW) and (GDGL) (Fig. 2).



Note: GC$^-$ - negative control groups, GDW - diabetic groups without treatment. GDGL - diabetic group + glibenclamide, GDES - diabetic group + the ethanolic extract of *S. officinalis*

Figure 2. Evolution of glycemia during experimentation (means ± SEM with n = 4)

Otherwise, regarding the results of biochemical assessment (Table 5), it was noted that there occurred a highly significant increase in urea, creatinine, cholesterol and TGP in the group (GDW) and group (GDGL) against the group of control group

Table 4. Evolution of experimental rats weight and food intake during experimentation

	Initial weight (g)	Final weight (g)	Weight Gain (g)	Food intake (g per week)
GC$^-$	304.25 ± 2.49	333.02 ± 4.01	+28.77	248.06 ± 12.05
GDW	309.6 ± 6.74	241.6 ± 12.88***	-68	275.55 ± 14.56###//#
GDGL	299.52 ± 5.43	195.1 ± 18.5***	-104.42	232.41 ± 25.75
GDES	287.66 ± 3.52	279.23 ± 28.61***	-8.43	237.25 ± 23.48

Note: Results are presented as means ± SEM with n = 4); GC$^-$: negative control groups; GDW - diabetic group without treatment; GDGL - diabetic group + glibenclamide; GDES - diabetic group + the ethanolic extract of *S. officinalis*.; *** - a highly significant difference between GC$^-$ vs GDW, GDGL and GDES; ### - a highly significant difference between GDW vs GDGL and GDES. # - a highly significant difference between GDW vs GC$^-$.

Table 5. Blood test results of experimental rats for antidiabetic activity (means ± SEM with n = 4)

	Urea	Creatinine	Cholesterol	Triglycerides	TGO	TGP
GC$^-$	43 ± 1.87	10.45 ± 0.45	61 ± 5.7	79.57 ± 10.84	100.18 ± 18.93	50.15 ± 5.75
GDW	121.75 ± 10.59***	18.6 ± 0.45***	97 ± 5.14***	86 ± 1.96	161.6 ± 20.02	81.76 ± 3.7***
GDGL	143.25 ± 22.5	18.83 ± 0.63	103.75 ± 8.75	81.5 ± 7.12	162.85 ± 19.08	89.26 ± 2.63
GDES	84 ± 18#	14.58 ± 2.02#	82.25 ± 6.32#	86.25 ± 10.92	145.15 ± 28.97	76.47 ± 5.48

Note: glycemia, urea, cholesterol and triglycerides in mg dL^{-1}. Creatinine in mg L^{-1} and TGO with TGP in IU/l.). GC$^-$ - negative control groups; GDW - diabetic group without treatment; GDGL - diabetic group + glibenclamide, GDES - diabetic group + the ethanolic extract of *S. officinalis*; *** - a highly significant difference between GC$^-$ vs GDW and GDGL; # - a significant difference between GDES vs GDGL and GDW.

GC$^-$. On the other hand, the GDES group reported a significant decrease in urea, creatinine and cholesterol compared to the GDW group. For triglycerides and TGO, a non-significant difference was shown between the four groups GC$^-$, GDW, GDGL and GDES.

Results presented an increase in blood glucose, urea, creatinine, cholesterol, triglycerides and TGP in diabetic rats. It can be explained by the administration of the effect of chemical diabetogen "Alloxan" which induced lipid, carbohydrate and protein damage accompanying oxidative stress applied to insulin-producing cells (Jayant and Srivastava, 2015). Previous studies showed that high urea and creatinine levels in diabetics indicated renal dysfunction caused by high protein breakdown. This led to an accumulation of amino acids in the serum and consequently to an increase in the concentration of serum enzymes (TGO and TGP) (Marella et al., 2015; Mirmohammadlu et al., 2015; Sirivole and Eturi, 2017; Zhang et al., 2017). In diabetics, the increase in cholesterol due to fatty acids released peripheral deposits in the absence of insulin action which was involved in the inhibition of fat storage in adipose tissue by the hormone-sensitive lipase (Vergès, 2001; Mariee et al., 2009; Hossein et al., 2013). At the same time, the increased concentration of serum enzymes (TGO and TGP) in the serum of diabetics can be explained by an exaggerated degradation of proteins leading to an accumulation of amino acids in the serum. This indicated liver damage due to a loss of membrane permeability (Hossein et al., 2013; Marella et al., 2015; Mirmohammadlu et al., 2015). Sharma et al. (2006) explained the productive power of free radicals due to hyperglycemia. Another study showed that the oxidative stress provided by hyperglycemia could lead to disturbances in the metabolism of proteins, lipids and carbohydrates (Jamaludin et al., 2016). The obtained results were fully consistent with the work of Eidi et al. (2009) who noted that the administration of ethanolic extract (80%) of *S. officinalis* caused a decrease in blood sugar, cholesterol, urea and creatinine levels. Aqueous and ethanolic extracts of sage caused a significant decrease in blood sugar, cholesterol and triglyceride levels (Ghowsi et al., 2019).

Khashan and Al-Khefaji (2015) exposed that aqueous and ethanolic extracts of sage at a dose of 100 mg kg^{-1} administered over a period of 14 days were capable of entering a significant reduction in blood sugar levels, cholesterol and triglyceride compared to the drug "Glibenclamide". In addition, several studies confirmed that consumption of sage tea led to a reduction in blood glucose and cholesterol (Bassil et al., 2015; Ghowsi et al., 2019). It has been known that *S. officinalis* has pharmacological properties including antioxidant, anti-inflammatory, antimicrobial, antidiabetic and hypolipidemic (Ghorbani and Esmaeilizadeh, 2017; Ben Khaeder et al., 2018). However, the hypoglycemic power of ethanolic extract of *S. officinalis* can be explained by its richness in bioactive compounds (Ghorbani and Esmaeilizadeh, 2017) known for their antidiabetic effect of polyphenols, flavonoids, tannins and alkaloids. In addition, flavonoids and tannins caused the reduction of blood glucose, cholesterol and triglyceride in diabetic rats (Velayutham et al., 2012; Obafemi et al., 2017). Flavonoids affected blood glucose levels, where they acted directly on pancreatic β- cells and through their regeneration and prevention of destruction led to increased insulin secretion or indirectly through inhibition of α-

glucosidase in the intestine and improved glycogen storage in the liver (Belmouhoub et al., 2017; Marella, 2017). On the other hand, alkaloids were also involved in this process by reducing glucose transport through the intestinal epithelium and improving liver glycogen content (Sani, 2015; Aba and Asuzu, 2018).

Conclusion

The richness of ethanolic extract of *Salvia officinalis* with bioactive compounds provided a major role in reducing hyperglycemia in diabetics at the same time stabilizing cholesterol, urea, creatinine, triglycerides and TGP compared to glibenclamide. As a result, it demonstrated the power of using ethanolic extract of *S. officinalis* as a natural medicine for the treatment of diabetes and its complications.

Funding

This research has not received any specific funding from public, commercial or not-for-profit funding agencies.

Acknowledgements

Authors would like to thank the engineers of Laboratory Reproduction of Farm Animals and Laboratory of Animal Hygiene and Pathology of Institute of Veterinary Sciences University Ibn Khaldoun Tiaret. Thanks also to the Directorate General of Scientific Research and Technological Development "DGRSDT".

References

Aba P. E., Asuzu I. U. (2018). Mechanisms of Actions of Some Bioactive Antidiabetic Principles from Phytochemicals of Medicinal Plants: A Review. Indian J Nat Prod Resour 9 (2): 85-96.

Alarcon-Aguilar F.J., Roman-Ramos R., Flores-Saenz J.L., Aguirre-Garcia F. (2002). Investigation on the Hypoglycemic Effects of Extracts of Four Mexican Medicinal Plants in Normal and Alloxan-Diabetic Mice. Phytother Res 16: 383–386. doi: 10.1002/ptr.914.

Bagre I., Bahi C., Gnahoue G., Djaman A.J., Guede G.F. (2007). Phytochemical Composition and *in vitro* Evaluation of the Antifungal Activity of Extracts of *Morinda morindoides* (Baker) Milne-Redhead (*rubiaceae*) Leaves on Aspergillus *fumigatus* and *candida albicans*. Science Pharm Biol 8 (1): 15-23.

Bassil M., Daher Costantine F., Mroueh M., Zeeni N. (2015). *Salvia libanotica* Improves Glycemia and Serum Lipid Profile in Rats Fed a high Fat Diet. BMC Complement Altern Med 15(384): 1-7. doi: 10.1186/s12906-015-0917-8.

Belmouhoub M., Chebout I. (2017). Antidiabetic and Antihypercholesterolemic Effects of Flavonoid-Rich Fractions of *Rosmarinus officinalis* in Streptozotocin-Induced Diabetes in Mice. Phytother 1-7. Doi :10.1007/s10298-017-1103-6.

Ben Khedher M.R., Hammami M., Arch Jonathan R.S., Hislop D.C., Eze D., Edward T.W., Małgorzata A.K., Zaibi M.S. (2018). Preventive Effects of *Salvia officinalis* Leaf Extract on Insulin Resistance and Inflammation in a Model of High Fat Diet Induced Obesity in Mice That Responds to Rosiglitazone. Peer J 6: 1-22. doi: 10.7717 /peerj.4166.

Ben Younes A., Ben Salem M., El Abed H., Jarraya R. (2018). Phytochemical Screening and Antidiabetic, Antihyperlipidemic and Antioxidant Properties of *Anthyllis henoniana* (Coss.) Flowers Extracts in an Alloxan-Induced Rats Model of Diabetes. Evid Based Complement Altern Med 1-14. doi: 10.1155/2018/8516302.

Bidie A., N'guessan Banga B., Yapo A. F., N'guessan J.D., Djaman A.J. (2011). Antioxidant Activities of Ten Medicinal Plants from the Ivorian Pharmacopoeia. Sci Nat 8 (1): 1 - 11.

Eidi A., Eidi M. (2009). Antidiabetic Effects of Sage (*Salvia officinalis* L.) Leaves in Normal and Streptozotocin-Induced Diabetic Rats. Diabetes Metab Syndr: Clin Res Rev 3: 40–44. doi: 10.1016/j.dsx.2008.10.007.

Eidi A., Eidi M., Shahmohammadi P., Mozaffarian V., Rustaiyan A., Mazooji A. (2011). Antidiabetic Effect of *Salvia verticillata* L. Aerial Parts in Normal and Streptozotocin-Induced Diabetic Rats. Inter J Pharmacol 7 (1): 66-73. doi:10.3923/Ijp.2011.66. 73.

El Ouali L. A., El-Akhal F., Ez-zoubi Y., Taghzouti K. (2016). Study of Phytochemical Screening and Larvicidal Efficacy of Ehtanolic Extract of *Salvia officinalis* (Lamiaceae) from North Center of Morocco against *Culex pipiens* (Diptera: Culicidae) Vector of Serious Human Diseases. Inter J Pharmacogn Phytochem Res 8(10): 1663-1668.

Ewenighi C. H., Dimkpa U., Onyeanusi J., Onoh L., Onoh G., Ezeugwu U. (2015). Estimation of Glucose Level and Body Weight in Alloxan Induced Diabetic Rat Treated with Aqueous Extract of *Garcinia Kola* Seed. Ulutas Med J 1 (2): 26-30. doi : 10.5455/umj.20150507042420.

Favier A. Stress oxydant et pathologies humaines [Oxidative stress in human diseases]. Ann Pharm Fr. 64 (6):390-396. doi: 10.1016/s0003-4509(06)75334-2. (in French)

Forbes J.M., Cooper M.E. (2013). Mechanisms of Diabetic Complications. Physiol Rev 93 (1):137-88. doi: 10.1152/physrev.00045.2011.

Frank B. H., Joann E. M., Meir J. S., Graham C., Simin L., Caren G. S., Walter C. W. (2001). Diet, Lifestyle and the Risk of Type 2 Diabetes Mellitus in Women. New Eng J Med 345 (11): 790-797.

Gebru A. K., Sibhat G. G., Hiben M. G. (2014). Preliminary Phytochemical Screening and Blood Glucose Lowering Activity of Methanol Extract of *Salvia tilifolia* Vahl Aerial Part. Int J Pharm Biol Arch 5(5): 122 – 125.

Ghezelbash G. R., Parishani M. R., Fouani M. H. (2015). Antimicrobial Activity of *Salvia officinalis* Acetone Extract against Pathogenic Isolates. Herb Drugs 5 (4): 215-218.

Ghorbani A., Esmaeilizadeh M. (2017). Pharmacological Properties of *Salvia officinalis* and Its Components. J Tradit Complement Med 2017: 1-8. doi: 10.1016/j.jtcme.2016.12.014.

Ghowsi M., Yousofvand N., Moradi S. (2019). Effects of *Salvia officinalis* L. (Common Sage) Leaves Tea on Insulin Resistance, Lipid Profile and Oxidative Stress in Rats with Polycystic Ovary: An Experimental Study. Avicenna J Phytomed 1-10.

Harborne J. B. (1998). Phytochemical Methods. A Guide to Modern Techniques of Plants Analysis. 3ème edition, Chapman et Hall, Britanie, p. 294.

Hartman R. E., Ross D. M. (2018). Effects and Mechanisms of Actions of Phytochemicals on Alzheimer's Disease Neuropathology. Frontiers In Biosci Elite 10:300-333.

Hossein A., Reza H., Vahid N., Minoo I. (2013). Effects of Aqueous Extract of *Berberis integerrima* Root on Some Physiological Parameters in Streptozotocin-Induced Diabetic Rats. Iranian J Pharm Res 12 (2): 425-434.

Jamaludin M., Nazratun N. A. H., Zariyantey A. H., Budin S.B. (2016). Mechanisms of Diabetes-Induced Liver Damage The Role of Oxidative Stress and Inflammation. Sultan Qaboos Univ Med J 16 (2):132–141. doi:10.18295/squmj.20 16.16.02. 002

Jayant Shiv K., Srivastava N. (2015). Evaluate Insulin Secretagogue Activity in Diabetic Rats by Aqueous Suspension of Garlic. Integr Obes Diabetes 2 (1): 183-186. doi: 10.15761/IOD.1000141.

Kadhim S. M., Mohammed M. T., Ahmed O. M., Jassimand A. M. N. (2016). Study of Some *Salvia officinalis* L. (Sage) Components and Effect of Their Aqueous Extract on Antioxidant. Int J Chem Sci 14(2): 711-719.

Kahn A. P. (2005). Diabetes: Causes, Prevention and Treatment. Orient Paperbacks, India, p. 24.

Khare R., Upmanyu N., Jha M. (2019). Exploring the Potential Effect of Methanolic Extract of *Salvia officinalis* against UV Exposed Skin Aging: *In vivo* and *In vitro* Model. Curr Aging Sci 12: 1-10. doi: 10.2174/1874609812666190808140549

Khashan K. T., Al-khefaji K. A. (2015). Effects of *Salvia officinalis* (L.) (Sage) Leaves Extracts in Normal and Alloxan-Induced Diabetes in White Rats. Inter J Sci Engineer Res 6 (1): 20-27.

Krimat S., Dob T., Toumi M., Kesouri A., Noasri A. (2015). Assessment of Phytochemicals, Antioxidant, Antimicrobial and Cytotoxic Properties of *Salvia chudaei* Batt. et Trab. Endemic Medicinal Plant from Algeria. J Mater Environ Sci 6 (1): 70-78.

Li X., Luo Y., Wang L., Li Y., Shi Y., Cui Y., Xue M. (2010). Acute and Subacute Toxicity of Ethanol Extracts from *Salvia przewalskii* Maxim in Rodents. J Ethnopharmacol 131: 110-115. doi: 10.1016/j.jep.2010.06.012

Marcelo S. P., luhara S. R., lEila S., Rafaianne Q.M.S., Thaigra S. S., Madileine F. A., Kleber E. C., Débora C. D., Gustavo T. V. (2017). Effect of *Bauhinia holophylla* Treatment in Streptozotocin-Induced Diabetic Rats. Anais da Academia Brasileira de Ciências 89 (1) : 263-272. doi: 10.1590/0001-3765201720160050

Marella S. (2017). Flavonoids-the Most Potent Poly-Phenols as Antidiabetic Agents: An Overview. Modern Approaches in Drug Designing 1(3):1-5. doi: 10.31031/MADD.20 17.01.000513.

Marella S., Dilip R. M., Kameswara R. B., Jyothi K. M. V., Chippad A. (2015). Antihyperlipidemic and Biochemical Activities of Mcy Protein in Streptozotocin- Induced Diabetic Rats. Cell Physiol Biochem 35: 1326-1334. doi: 10. 1159/000373954.

Mariee A. D., Abd-Allah G. M., El-Yamany M. F. (2009). Renal Oxidative Stress and Nitric Oxide Production in Streptozotocin-Induced Diabetic Nephropathy in Rats: The Possible Modulatory Effects of Garlic (*Allium sativum* L.). Biotech App Biochem 52: 227–232. doi:10.1042/BA20080086.

Mirmohammadlu M., Hosseini S. H., Kamalinejad M., Gavgani M. E., Noubarani M., Eskandari M. R. (2015). Hypolipidemic, Hepatoprotective and Renoprotective Effects of *Cydonia Oblonga* Mill. Fruit in Streptozotocin-Induced Diabetic Rats. Iranian J Pharm Res 14 (4): 1207-1214.

Mujeeb F., Bajpai P., Pathak N. (2014). Phytochemical Evaluation, Antimicrobial Activity and Determination of Bioactive Components from Leaves of Aegle Marmelos. Biomed Res Inter 2014: 1-11. Doi: 10.1155/2014/497606.

Noudogbessi J. P. A., Natta A. K., Tchobo F. P., Bogninou G. S., Bothon F. T. D., Bossou A. D., Figueredo G., Chalard P., Chalchat J. C., Sohounhloué D. C. K. (2013). Phytochemical Screening of *Pentadesma butyracea* Sabine (Clusiaceae) Acclimated in Benin by GC/MS. Int Sch Res Notices 2013: 1-8. doi: 10.1155/2013/172397.

Obafemi T. O., Akinmoladun A. C., Olaleye M. T., Agboade S. O., Onasanya A. A. (2017). Antidiabetic Potential of Methanolic and Flavonoid-Rich Leaf Extracts of *Synsepalum Dulcificum* in Type 2 Diabetic Rats. J Ayurveda Integr Med 8:238-246. doi: 10.1016/j.jaim.2017.01.008

Oliveira G. O., Braga C. P., Fernandes A. A. H. (2013). Improvement of Biochemical Parameters in Type 1 Diabetic Rats after the Roots Aqueous Extract of Yacon [*Smallanthus sonchifolius* (Poepp. & Endl.)] Treatment. Food Chem Toxicol 59: 256–260. doi: 10.1016/j.fct.2013.05.050

Pratima H., Policepatil V. (2017). Phytochemical Screening and Antimicrobial Activity of Various Extracts of *Salvia aegyptiaca* L. Int J Curr Microbiol Appl Sci 1: 600-608. doi: 10.20546/ijcmas.2017.601.073.

Raju M. G., Sundar R. T. H. (2017). *Aristolochia bracteolata* – Antihyperglycemic and Antihyperlipidemic Activity in Dexamethasone-Induced Diabetic Rat Model. Asian J Pharm Clin Res 10(8): 75-77. doi: 10.22159/ajpcr. 2017.v10i8.18328

Raman R., Gella L., Srinivasan S., Sharma T. (2016). Diabetic Retinopathy: An Epidemic at Home and around the World. Indian J Ophthalmol, 64(1): 69–75. doi: 10.4103/0301-4738.178150

Ramırez J. H., Palacios M., Tamayo O., Jaramillo R., Gutierrez O. (2007). Acute and Subacute Toxicity of *Salvia scutellarioides* in Mice and Rats. J Ethnopharmacol 109: 348–353. doi: 10.1016/j.jep.2006.07.040

Sani U. M. (2015). Phytochemical Screening and Antidiabetic Effect of Extracts of the Seeds of *Citrullus lanatus* in Alloxan-Induced Diabetic Albino Mice. J App Pharm Sci 5 (03): 051-054. doi: 10.7324/JAPS.2015.50309

Sharma S., Kulkarni S. K., Chopra K. (2006). Curcumin, The Active Principle of Turmeric (*Curcuma longa*), Ameliorates Diabetic Nephropathy in Rats. Clin Exper Pharmacol Physiol 33: 940–945. doi: 10.1111/j.1440-1681.2006.04468.x.

Sirivole M. R., Eturi S. (2017). A Study on Blood Urea and Serum Creatinine in Diabetes Mellitus from Sangareddy District, Telangana, India. Int J Med Health Res 3 (12): 132-136.

Soni A., Sosa S. (2013). Phytochemical Analysis and Free Radical Scavenging Potential of Herbal and Medicinal Plant Extracts. J Pharmacogn Phytochem 2 (4): 22-29.

Tiwari P., Kumar B., Kaur M., Kaur G., Kaur H. (2011). Phytochemical Screening and Extraction: A Review. Inter Pharm Sci 1 (1): 98-106.

Velayutham R., Sankaradoss N., Ahamed K. F. (2012). Protective effect of tannins from Ficus racemosa in hypercholesterolemia and diabetes induced vascular tissue damage in rats. Asian Pac J Trop Med. 5 (5): 367-73. doi: 10.1016/S1995-7645(12)60061-3

Vergès B. (2001). Insulin-Sensibility and Lipids. Diabetes Metabolism 27 :223-227.

Zhang S., Xu H., Yu X., Wu Y., Sui D. (2017). Metformin Ameliorates Diabetic Nephropathy in a Rat Model of Low-Dose Streptozotocin-Induced Diabetes. Exper Therap Med 14: 383-390. doi: 10.3892/etm.2017.4475.

aCS88_40