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Research Article

Anti-hyperglycemic Effect and Liver Enzymes Activity of *Varthemia iphionoides* essential Oil in Diabetic Rats.

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ABSTRACT

The major goal of this study was to evaluate the anti-hyperglycemic effect and the activity of hepatic enzymes after treating the streptozotocin-induced diabetic rats with the essential oil of *Varthemia iphionoides* . 24 wistar albino rats weighing between 180-200g were randomly grouped into four categories of six rats. Group A, normal control was received distilled water only. Group B, negative diabetic control that consist if six rats induced with streptozotocin. Diabetic group (C) contains Metformin treated rats at concentrations of 100mg/kg. Diabetic group (D) orally treated with of *V. iphionoides* essential oil at concentrations of 200mg/kg of body weight. The treatment was done daily for two weeks. Transaminase as well anti-diabetic activity assessed by considering level and component of blood d plasma and glucose respectively. This work was conducted at the department of Biology, Yarmouk University, Irbid, Jordan, November, 2017. glucometeric as well as spectrophotometric approaches to quantify level of glucose and hepatic enzymes respectively. Results obtained at this study showed a significant reductions in glucose level in rats treated with *V.iphionoides* essential oil ($P<0.05$) compared to that of the diabetic rats. Significant reduction in AST level ($P<0.05$) was also obtained in group treated with *V. iphionoides* essential oil and metformin compared with diabetic group .On other hand no significant change in the level of ALP and ALT were noticed between the controls and the treated groups. Conclusion: the result possessed that *V. iphionoides* essential oil has anti-hyperglycemic activity and therefore safe and shares Metformin same effect on the hepatic enzymes.

Key words: Anti-Diabetic. *Varthemia iphionoides.* Liver Enzymes. Metformin. streptozotocin.

INTRODUCTION

Diabetes mellitus has been considered as major chronic disorders that characterized by abnormalities in the carbohydrate metabolism that lead to elevated in blood sugar (Ceriellos and Mortz, 2004). The elevated level of blood sugar is caused by defects in insulin production or function which usually cause chronic hyperglycaemia. The elevated glucose level is widely accepted as major stimuli for β-cells apoptosis (Hui *et al* 2004). Diabetes leads to many complications such as heart disease, kidney failure, foot problem, blindness, neuropathy and thickening of blood vessels that leading to cardiovascular diseases. It has been reported that the number of patients

suffering from diabetes in 2014 was 422 million by (Dhasarathan and Theriappan, 2011, WHO, 2016).

Different methods were used to induce diabetes mellitus experimentally, streptozotocin (STZ) is one of the most used chemical for this purpose. The mechanism action of STZ is effected by reactive oxygen species. It enters the β cell by GLUT2 and stimulates DNA alkylation. For diabetogenicity of streptozotocin the DNA disorder results in activation of poly ADP-ribosylation which is more important than DNA damage itself. Depletion of high energy signals including NAD+ as well as ATP arise as a result of Poly ADP-ribosylation. After streptozotocin treatment, ATP dephosphorylation is highly activated providing a substrate for the enzyme xanthine oxidase which as a result leads to the accumulation of superoxide radicals. Nitric oxide secreted at a toxic amounts due to the streptozotocin effect of that inhibit aconitase activity which finally damage DNA structure. Cells necrosis occurs as a result of streptozotocin action (Szkudelski T. 2001).

The liver is the largest internal organ and highly specialized tissue which detoxifies various metabolites, produces biochemical and synthesizes macromolecules as proteins. It also plays a role in metabolism, control of glycogen storage, degeneration of RBC and hormone production (Aja et al. 2015a).

It is therefore obvious that any disorder condition, which affects the liver cells, will leads to metabolic derangement (Szkudelski T. 2001).When hepatocytes damage occurs; blood stream was reported to have elevated hepatic enzymes. Medication in general and herbal extract in particular cause elevation of the hepatic enzymes, therefore the effect of any medication or herbal extract on the serum changes of the hepatic enzymes so important to ensure the hepato-protectiveness of such extract or drug. This can be controlled by liver function tests: alanine aminotransferase (ALT), alkaline phosphatase (ALP), as well as aspartate aminotransferase (AST) (Ochei and Kolhatkar, 2007).

Diabetes of is a world-wide problem and an efficient drug has yet to be identified (Dieye et al, 2008) .Metformin (Met) is the a widely used and accepted drug used to treat patients with Type- 2 diabetes (Kooy et al.2009). The major effect of Metformin is to reduce liver glucose production (Cheng et al, 2006). In addition, metformin reduce sugar absorption in the gastrointestinal tract, increases glucose entrance in liver and muscle cells, and reduce the level of serum free fatty acid by its ability to effect as anti-lipolytic and as a result substrate availability for gluconeogenesis is highly limited (Pournaghi et al, 2012).

Plants have long been used as a major source of both traditional and modern medicines. Herbal medicine had been used by 80% of populations as an efficient method for the treatment of diabetes mellitus. (Bamidele et al, 2014).

World Health Organization has recently estimated that more than 30 million patient worldwide suffering from diabetes mellitus use anti-diabetic herbs in effectively to treatment the disease (WHO, 2014). Most of the used anti-diabetic plants contain potentially useful constituents such as terpenoids, alkaloids, glycosides, and flavon that form the basic components for the manufacturing of modern pharmaceuticals (Aja et al, 2015b).

Varthemia iphionoides is one of the most common plant species known to treat diabetes. It has a long been used as an anti-diabetic herb. However, attempt to obtain active not only but lead to contrasting consequences. Reports show that Varthemia iphionoides extract possessed a moderate effect in fasting plasma glucose concentrations reduction in diabetic patients with poor glycemic control (Afifi et al, 1997).

MATRIALS AND METHODS

Collection of Plant Materials: The aerial parts of fully grown V. iphionoides were collected from Rehab city- north of Jordan at July 2017. The collected plant was identified by a taxonomist, Prof. Sawsan Oran of the department of Biology,

University of Jordan. A voucher specimen was deposited at the herbarium in the Department of Biology, Al-albayt University, Jordan.

Essential Oil Extraction: The aerial parts of V. iphionoides were subjected to steam distillation for 3.0 to 4.0h. The collected essential oils were decanted and dried over anhydrous sodium sulfate, then stored in refrigerator until used.

Experimental Design: Twenty four laboratory rats of Wistar albino with weight range between 180-200g were obtained from Yarmouk University, department of biological sciences animal house. Rats were initially acclimatized in cages for at least 10 days under appropriate laboratory conditions. After that animals were subdivided into four groups each contain six rats, then the animals were housed in separate cages. The grouping was done as follows:

Group A, normal control (non-diabetic rats).

Group B, negative diabetic control were induced with Streptozotocin

Group C, diabetic rats treated with Metformin at dose 100mg/kg B.Wt.

Group D, diabetic rats treated orally by gavage with 100 mg/Kg B.Wt of V. iphionoides essential oil. The treatment was repeated twice a day for two weeks.

Induction procedure: To induce diabetes Streptozotocin were dissolved in distilled water to get 100mg/ml concentration. Rats in groups B, C and D were initially weighed and then Streptozotocin was injected intra-peritoneally at 60 mg/kg of body weight using insulin syringes. Symptoms including polydipsia, polyuria, glycosuria were initially used to determine the incidence with Diabetes Mellitus 72hrs after rats treatment. Furthermore, concentration of blood glucose obtained from the veins of rats tails were also tested using glucometer to confirm diabetes (Rees and Alcolado, 2005). Rats with blood glucose level greater than 200mg/dl were considered to be diabetic (Grover et al, 2002).

Collection and Preparation of Blood: After 14 days of treatment, the rats were sacrificed by cervical dislocation under mild chloroform anesthesia and blood samples were collected by ocular punctures into heparinized tubes, and transferred into plain centrifuge tubes. Within 1 hour of collection the blood samples were centrifuged for 10min at 4000xg on a centrifuge to separate sera.

Blood Glucose Level Determination: Glucose oxidase method (GOM) was used to determine the level of blood sugar via single touch glucometer and reactive strips.

Determination of Liver Enzymes: Alkaline phosphatase (ALP), Alanine aminotransferase (ALT) and Aspartate aminotransferase (AST) were measured using commercial kits.

Data Analysis: Results were expressed as mean standard deviation. The one-way analysis of variance (ANOVA) was used to analyze the data followed by post-hoc tests. The results are considered significant at $P<0.05$.

RESULTS

Figure 1 show the effect of *V. iphionoides* essential oil on glucose level on streptozotocin-induced diabetic albino rats. The results possessed significant glucose level decrease ($P<0.05$) in the treated rats groups comparing with STZ diabetic rats.

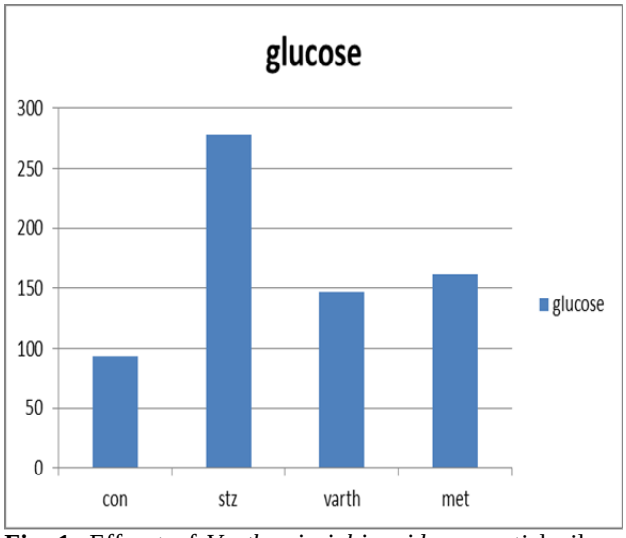

Fig. 1: Effacct of *Varthemia iphionoides* essential oil on Glucose level in diabetic rats induced with streptozotocin.

The effect of *Varthemia iphionoides* essential oil on liver enzymes in streptozotocin-Induced Diabetic Albino Rats:

To get more pieces of evidence for the efficient role of *V. iphionoides* essential oil on treating diabetes mellitus activity of hepatic enzymes of *V. iphionoides* essential oil treated rats was compared with that of the other groups. Figure 2 shows a significant decline ($P<0.05$) in the AST levels in treated groups compared to that of the other diabetic groups. Whereas a significant reduction ($P<0.05$) in the ALP levels was observed in rats streptozotocin-induced comparing to that of normal group. No significant ($P>0.05$) change in the levels of ALT was observed beteween diabetic rats and healthy groups.

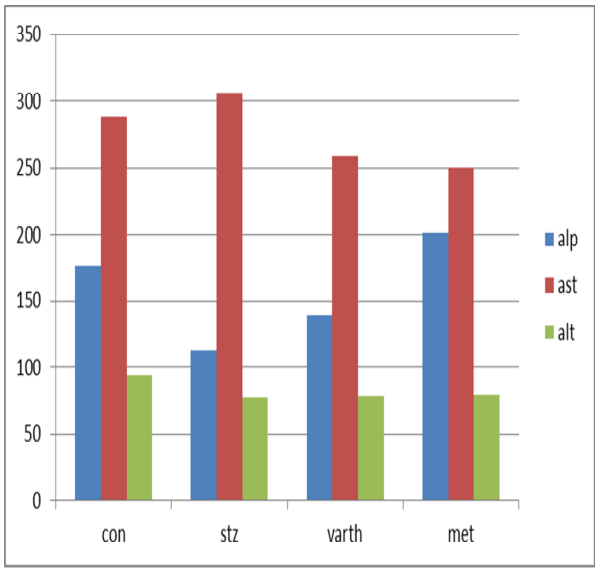

Fig. 2: Liver enzymes level in streptozotocin-induced diabetic rats treated with *V. iphionoides* essential oil.

DISCUSSION

Diabetes is a chronic disorder of carbohydrates metabolism which characterized by continuous elevation of blood glucose concentration and it is a large effects on worldwide (Grover *et al*, 2002). Due to the high cost of commercial drugs and the side effect, many researchers focused in the using of alternative treatment using herbal drugs. It has been estimated that about 800 plants species are involved to treat diabetes (Eidi *et al*, 2005) One such plant species that has recently been the subject of active research is *V. iphionoides*.

In this study, our result showed blood glucose level increase gradually the blood obtained from diabetic control group. That change was found to be significant ($P<0.05$) comparing to the normal control group. The intra-peritoneal induction of streptozotocin in the animals revealed significant ($P<0.05$) elevation in glucose level as earlier reported by Kazemian *et al*. (2015), they observed that after 72 hours of injection of streptozotocin dissolved in distilled water at 60mg/kg dose of body weight the glucose level of the rats were significantly ($P<0.05$) increased.

The capacity of *V. iphionoides* essential oil lower elevated blood glucose concentration significantly ($P < 0.05$) within normal limits, it is considered important for the liver to normalize its normal homeostasis in experimental diabetic rats. *V. iphionoides* essential oil may effects by activating the function of β-cells of pancreas in glucose metabolism as the result possessed in fig. 1. It is may have an unidentified factor that made normalization of blood glucose level and improve function of pancreas cells in glucose metabolism. Results of studies done by Afifi and colleagues revealed that the administration of *V. iphionoides* essential oil as gavage to diabetic marked with strptozotocin administration, reduced blood glucose levels in hyperglycemic and control groups. The decreasing of blood glucose level by *V. iphionoides* essential oil may be due to its numerous bioactive compounds such as phenols, flavonoids, saponins, triterpenoids and tannins,these compounds may play their role in hypoglycemic effects by increasing release and decreasing glucagon's secretion, decreasing insulin resistance, slowing the absorption of glucose or by reducing hepatic glucose production (Afifi et al. 1997).

The elevation of AST enzymes in plasma may be explained partially as a result of transporting of this enzyme from the liver cells into the bloodstream, which caused by hepatotoxic effects of streptozotocin (Zarei *at el*, 2015). *V. iphionoides* essential oil significantly decrease

($P<0.05$) the levels of AST in diabetic group comparing to normal control group (Figure 2). Administration of the *V. iphionoides* essential oil reduced AST serum level which may be related to a decrease in ATP production in the absence or shortage of insulin. This may be related to decreased levels of plasma proteins in streptozotocin induced rats, no significant ($P>0.05$) reductions was observed in treated groups comparing to diabetic groups.

CONCLUSION

Data obtained in this piece of work clearly suggests that *V. iphionoides* essential oil has anti-hyperglycemic effect on streptozotocin-induced diabetic albino rats. Like metformin, *V. iphionoides* essential oil has the same effect on liver enzymes and therfor concluded to be relatively safe and may reduce the liver damage induced by streptozotocin.

AKNOLEDGMENT

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REFERENCES

1. Afifi, F., Saket, M. Jaghabir, M. and Al-Eisawi, D. 1997. Effect of *Varthemia iphionoides* on blood glucose level of normal rats and rats with streptozocin-induced diabetes mellitus. Current Therapeutic Research. 58 (11). 888-892.
2. Aja, P.M. Ani, O.G. Offor, C.E. Orji, U.O. and Alum, E.U.. 2015a.Evaluation of Anti-Diabetic Effect and Liver Enzymes Activity of Ethanol Extract of *Pterocarpus santalinoides* in Alloxan Induced Diabetic Albino Rats. Global Journal of Biotechnology & Biochemistry 10 (2): 77-83.
3. Aja, P.M. Igwenyi, I.O. Ugwu Okechukwu P.C., Orji O.U. and Alum, E.U.. 2015b. Evaluation of Anti-diabetic Effect and Liver Function Indices of Ethanol extracts of *Moringa oleifera* and *Cajanus cajan* Leaves in Alloxan Induced Diabetic Albino Rats. Global Veterinaria 14 (3): 439-447.
4. Bamidele, O., D.S. Arokoyo, A.M. Akinnuga and A.O. Oluwarole, 2014. Anti-diabetic Effect of Aqueous Extract of *Basella alba* Leaves and Metformin in Alloxan Induced Diabetic Albino Rats. African Journal of Biotechnology, 13(24): 2455-2458.
5. Ceriellos, A. and Mortz, E. 2004. Is Oxidative Stress the Pathogenic Mechanism underlying Insulin Resistance, Diabetes and Cardiovascular Disease? Arteriosclerosis Thrombosis and Vascular Biology, 24: 816-823.
6. Cheng, J.T., C.C. Huang, I.M. Liu, T.F. Tzeng and C.J. Chang, 2006. Novel Mechanism for Plasma Glucose Lowering Action of Metformin in Streptozotocin-induced Diabetic Rats. Diabetes Care, 55: 819-825.
7. Dhasarathan, P. and P. Theriappan, 2011. Evaluation of Anti-diabetic Activity of *Strychonous potatorum* in Alloxan-induced Diabetic Rats. Journal of Medicine and Medical Sciences, 2(2): 670-674.
8. Dieye, A.M., A. Sarr, S.N. Diop, M. Ndiaye, G.Y. Sy and M. Diarra, 2008. Medicinal Plants and the Treatment of Diabetes in Senegal: Survey with Patients. Fundamental and Clinical Pharmacology, 22: 211-216.
9. Eidi , M. Eidi A., Zamaniza , H. 2005. Effect of Salvia officinalis L. leaves on
10. serum glucose and insulin in healthy and streptozotocin-induced diabetic rats. Journal of Ethnopharmacology, 100: 310–313.
11. Grover,J. K. Yadav, S. Vats, V. 2002. Medicinal plants of India with anti-diabetic potential. Journal of Ethnopharmacology, 81: 81-100.
12. Kazemian Mansur Abad M, Haeri MR, Ebrahimi M , Heidari R. 2015. Anti-diabetic effect of *Capparis spinosa* L. root extract in diabetic rats. Avicenna J Phytomed, 5 (4): 325-332.

13. Kooy, A., J. De Jager, P. Lehert, D. Bets, M.G. Wulffele and A.J.M. Donker, 2009. Long-term Effects of Metformin on Metabolism and Micro-vascular and Macro-vascular Disease in Patients with Type 2 Diabetes Mellitus. Archives of International Medicine, 169: 616-625.
14. Ochei, J. and A. Kolhatkar, 2007. Medical Laboratory Science: Theory and Practice. 6th Edition,Tata McGraw Hill, New Delhi, pp: 106-158.
15. Pournaghi, P., Sadrkhanlou R., Hasanzadeh S. and Foroughi, A. 2012. An Investigation on Body Weights, Blood Glucose Levels and Pituitary-gonadal Axis Hormones in Diabetic and Metformin-treated Diabetic Female Rats. Veterinary Research Forum, 3(2): 79-84.
16. Rees, D.A. and J.C. Alcolado, 2005. Animal Models of Diabetes Mellitus. Diabetic Medicine, 22: 359-70.
17. Szkudelski T. 2001.The mechanism of alloxan and streptozotocin action in B cells of the rat pancreas. Physiol Res.; 50(6):537-46
18. WHO., 2014. Diabetes mellitus. Report of a WHO Study Group. Geneva, World Health Organization Technical Report Series, pp: 727.
19. WHO., 2016. Diabetes mellitus. Report of a WHO Study Group. Geneva, World Health Organization Technical Report Series, pp: 6.
20. Zarei A,Vaezi GH, Malekirad AA, Abdollahi M. Effects of ethanol extract of *Salvia hydrangea* on hepatic and renal functions of streptozotocin-induced diabetic rat. Avicenna J Phytomed, 2015; 5 (2): 138-147.