SUMMARY AND CONCLUSIONS

DM is a disease marked by high levels of blood glucose resulting from a defect in insulin production, insulin action or both, that can lead to macro and micro vascular complications. It is a chronic, life threatening condition that depends on medication, diet and life style modification to prevent long term complications. If the imbalanced of glucose homeostasis does not return to normal and continues for a protracted period of time, it leads to hyperglycemia that is due course turns into a syndrome called DM.

Panax ginseng is a popular herbal remedy that has been used for thousands of years. There are five main species of ginseng: American, Chinese, Korean, Japanese and Siberian (or Russian).

This herb has a wide base of application and is considered the most popular herbal medicine worldwide. It has been used to treat a variety of disorders including: anaemia, insomnia, dyspnea, memory impairment, confusion, decreased libido, chronic fatigue, angina and DM.

CYP2E1 is a member of the CYP450 super family and primarily involved in Phase I metabolism where its major role is detoxification and bioactivation of drugs and xenobiotics.

So the aim of the study is to investigate the effect of *Panex ginseng* extract on CYP2E1 mRNA gene expression, some oxidative stress marker (MDA) and some antioxidant parameters (GSH and GPx) in STZ-induced diabetic rats.

This study was carried out on 50 rats which divided into five groups as follows:

Group I : 10 rats were served as normal controls (control group).

Group II : 10 diabetic rats (diabetes was induced by a single intraperitoneal injection of STZ at a dose of 60 mg/kg body weight for 3 consecutive days) (diabetic group).

Group III : 10 rats were received daily *Panax ginseng* extract 4 % dissolved in distilled water, (administered orally at a dose of 100 mg/kg body weight for 30 days) (ginseng group).

Group IV : 10 rats were pretreated with *Panax ginseng* extract as in group III for 30 days. Then they were injected by STZ as in group II for 3 consecutive days (ginseng-pretreated diabetic group).

Group V : 10 diabetic rats as in group II, then they were treated with *Panax ginseng* extract as in group III for 30 days (ginseng-treated diabetic group).

When comparing the final weight of rats with its corresponding initial weight it is found that, the body weight was significantly increased in the control, the ginseng and the ginseng-pretreated diabetic groups and significantly decreased in the diabetic and the ginseng-treated diabetic groups. As compared to their corresponding value of the control group: the final weight of rats was significantly decreased in the diabetic group, while significantly increased in the ginseng group and in groups IV and V it didn't show any significant difference. As compared to group II: the final body weight was significantly increased in group III, while in groups IV and V it didn't show any significant difference.

The following determinations were done for all the studied groups: (ALT, AST and GPx) activities and (FPG, Cholesterol, TG, GSH, MDA and CYP2E1 mRNA gene expression) levels as well as the histopathological examination of liver and pancreas.

Our results revealed that:

1- **In the diabetic group**: The values of FPG, Cholesterol, TG, ALT, AST, MDA and CYP2E1 mRNA gene expression were significantly increased as compared to their corresponding values of the control group, while GSH content and GPx activity were significantly decreased.

2- **In the ginseng group :** The values of FPG, Cholesterol, TG, ALT, AST, MDA,GSH,GPx and CYP2E1 mRNA gene expression didn't show any significant difference when compared to their corresponding values of the control group, while they were significantly decreased as compared to their corresponding values of the diabetic group except GSH content and GPx activity were significantly increased.

3- **In both the ginseng-pretreated diabetic group and the ginseng-treated diabetic group :** The values of FPG, Cholesterol, TG, ALT, AST, MDA and CYP2E1 mRNA gene expression were significantly increased as compared to their corresponding values of the control group, while GSH content and GPx activity were significantly decreased. While, all parameters were significantly decreased as compared with diabetic group except GSH content and GPx activity were significantly increased.

The histopathological study:

1- **In the control group**: The liver showed normal histological appearance. The pancreas showed normal islet of langerhans and normal acini tissues, islets were regular with well defined boundaries.

2- **In the diabetic group:** The liver showed degeneration in the surrounding hepatocytes and reduction in the number of nuclei, and inflammatory cells infiltrations were seen. The pancreas showed reduction in the number of cells in the islet, signs of necrosis in the islet, and severe reduction in the number of cells.

3- **In the ginseng group:** The liver showed revealing normal architecture, with regenerating hepatocytes. The pancreas showed islet cells and acini were look like normal cells.

4- **In the ginseng-pretreated diabetic group:** The liver showed nearly normal histological appearance, in spite it was accompanied by vacuolization in some cells and pyknotic nuclei were still observed. The pancreas showed nearly normal islets of langerhans while degeneration of some cells was still observed.

5- **In the ginseng-treated diabetic group:** the liver showed revealed nearly normal restoration of hepatocytes and sinusoids. Although dilatation in sinusoids were observed. The pancreas showed revealed remarkable improvement in the islet of langerhans. There was an increase in the islet cellular density.

There were a significant positive correlation between CYP2E1 mRNA gene expression and each of ALT activity, AST activity, FPG, Cholesterol, TG and MDA levels. While CYP2E1 mRNA gene expression demonstrated a negative significant correlation with GSH content and GPx activity.

From the results obtained in this study, we can conclude that:

- Oral administration of ginseng extract to diabetic rats exhibited anti diabetic, hepatoprotective, anti hyperlipidemic and antioxidants activities. These results suggest the possibility of using ginseng plant to treat patients who suffer from DM due to its good anti diabetic effect.

- The mean values of fasting blood sugar, liver function test, antioxidants parameters. CYP2E1 mRNA gene expression were not changed in ginseng group as compared to the control group, indicating its safety.

- The therapeutic effect of ginseng is more pronounced than the protective effect.

- The use of newly developed real-time quantitative reverse transcription polymerase chain reaction may improve the effectiveness of determination the expression of CYP2E1 mRNA gene expression.

RECOMMENDATIONS

1- Further research and development will be recommended to combine ginseng with other liver drugs to investigate their possible synergic efficacy and perferably pharmacological properties.

2- Ginseng and its principle components ginsenosides have shown benficial activities including important role in the regulation of liver functions and the treatment of the liver disorders of acute / chronic hepatotoxicity, hepatites, hepatic fibrosis / cirrhosis, even liver failure and hepatocelluer carcinoma. So the possible activity pathways of the actions have been also investigated. More detailed molecular mechanisms of activities of ginseng / ginsenosides as well as further efficacy and safety studies remain to the explosed.

REFERENCES

1- Shaw JE, Sicree RA, Zimmet PZ. Global estimates of the prevalence of diabetes for 2010 and 2030. Diabetes Research and Clinical Practice 2010; 87: 4-14.

2- Hashmi NR, Manzoor I, Daud S. Diabetes Mellitus; awareness among individuals attending out patient department of Ghurki Trust Teaching Hospital Professional. Medical Journal 2008; 15: 96-100.

3- Tiwari Ak, Rao JM. Diabetes mellitus and multiple therapeutic approaches of phytochemicals: Present status and future prospects. Current science 2002; 83(1): 30- 8.

4- Sollu M, Banji D, Otilia J, Banji F, Vijayalaxmi C. Srilath K, et al. Appraisal on causes and treatment of Diabetes Mellitus. Archives of Applied Science Research, 2010, 2 (5):239-60.

5- Shen GX. Lipid Disorders in Diabetes Mellitus and Current Management. Current Pharmaceutical Analysis 2007; 3: 17-24.

6 – Bastaki S. Diabetes mellitus and its treatment. International Journal of Diabetes &Metabolism 2005; 13: 111-34.

7- Chou KC. Molecular Therapeutic Target for Type-2 Diabetes. Journal of Proteome Research 2004; 3: 1284-8.

8- Smith M, Hopkins D, Peveler RC, Holt RIG, Woodward M, Ismail K. First v.second-generation antipsychotics and risk for diabetes in schizophrenia: systematic review and meta-analysis. The British Journal of Psychiatry 2008; 192: 406–11.

9 – Ahmadi E, Rahnama Z, Tehrani AR. atopic dermatitis and type 1 diabetes mellitus in Iranian children.American Journal of Immunology 2009; 5: 98-100.

10- Dejkhamron P, Menon RK, Sperling MA. Childhood diabetes mellitus: Recent advances & future prospects. Indian Journal of Medical Research 2007; 125: 231-50.

11- Bajaj JS. Management of Diabetes Mellitus: Principles and Practice. Kuwait Medical Journal 2002; 34: 94-105.

12- Wong LY, Toh MM. Understanding of Diabetes Mellitus and Health-preventive Behaviour Among Singaporeans. Ann Acad Med Singapore 2009; 38: 478-86.

13- Tuomilehto J, Lindstrom J, Eriksson JG, Valle TT, Hamalainen H, Parikka PI, et al . Prevention of type 2 diabetes mellitus by changes in life style amonge subjects with impaired glucose tolerance . The New England Journal of Medicine 2001; 344: 1343-50.

14- Ahmed N, Khan J, Siddiqui TS. Frequency of dyslipidaemia in type 2 diabetes mellitus in patients of Hazara division. Journal of Ayub Medical College Abbottabad 2008; 20: 51-4.

15- Eckardstein AV, Schulte H, Assmann G. Risk for Diabetes Mellitus in Middle-Aged Caucasian Male Participants of the PROCAM Study: Implications for the Definition of Impaired Fasting Glucose by the American Diabetes Association. The Journal of Clinical Endocrinology & Metabolism 2000; 85(9): 3101-8.

16 – Srinivasan BT, Jarvis J, Khunti K, Davies MJ. Recent advances in the management of type 2 diabetes mellitus: a review. Postgraduate Medical Journal 2008; 84: 524–31.

17- Vijan S, Rodney A. Treatment of Hypertension in Type 2 Diabetes Mellitus: Blood Pressure Goals, Choice of Agents, and Setting Priorities in Diabetes Care Hayward. Annals of Internal Medicine 2003; 138: 593-602.

18- Chan JL, Abrahamson MJ. Pharmacological management of type 2 diabetes : rationale for rational use of insulin. Mayo Clinic Proceedings 2003; 78: 459-67.

19- Moeini SH, Mirmiran P, Mehrabi Y, Azizi F. Evaluation of different risk factors for early diagnosis ofdiabetes mellitus.Iran Journal of Medical Sciences 2004; 29: 21-5.

20- Jimenez-Moleon JJ, Bueno-Cavanillas A, Luna-del-Castillo1 JD, Garcia-Martin M, Lardelli-Claret P, Galvez-Vargas R. Prevalence of gestational diabetes mellitus: variations related to screening strategy used. European Journal of Endocrinology 2002; 146: 831–7.

21- Chen Y, Quick WW, Yang W, Zhang Y, Baldwin A, Moran J, et al. Cost of Gestational Diabetes Mellitus in the United States in 2007. Population health management 2009 ; 12: 165-74.

22- Kumar SN. Gestational diabetes mellitus. KMJ 2009; 3: 115-9.

23- Mealey BL, Ocampo GL. Diabetes mellitus and periodontal disease. Periodontology 2007; 44: 127–53.

24- Etuk EU, Muhammed BJ. Evidence based of chemical method of induction of diabetes mellitus in experimental animals. Asian Journal of Expetrimenal Biological Sciences 2010; 1(2):331-6.

25- Agarwal DK, Jeloka T, Sharma AP, Sharma RK. Steroid induced diabetes mellitus presenting as diabetic ketoacidosis. Indian Journal of Nephrology 2002; 12:122-3.

26- Alberti KG, Zimmet PZ. Definition, Diagnosis and Classification of diabetes mellitus and its complications.Part1: Diagnosis and Classification of diabetes mellitus provisional report of a WHO consultation. Diabet Med 1998; 15:539-53.

27- American Diabetes Association. Diagnosis and Classificationof diabetes mellitus. Position statement. Diabetes Care 2005; 29(1):37-42.

28 - Virtue MA, Furne JK, Nuttall FQ, Levitt MD. Relationship between GHb concentration and erythrocyte survival determined from breath carbon monoxide concentration. Diabetes Care 2004; 27:931-5.

29- Rohlfing CL, Wiedmeyer HM, Little RR, England JD, Tennill A, Goldstein DE. Defining the relationship between plasma glucose and HbA1c: Analysis of glucose profiles and HbA1c in the Diabetes Control and Complications Trial. Diabetes Care 2002; 25:275-8.

30- Davidson MB, Schriger DL, Peters AL, Lorber B. Glycosylated hemoglobin as a diagnostic test for type 2 diabetes mellitus. JAMA 2000; 283(5):605-7.

31- Shukla R, Sharma SB, Puri D, Murthy PS. Indigenous Drugs for Diabetes Mellitus. J K Science 2000; 2: 128-30.

32- Tiwari AK, Rao JM. Diabetes mellitus and multiple therapeutic approaches of phytochemicals: Present status and future prospects. Current science 2002; 83: 30-8.

33- Rakesh KV, Kumar V, Kothari A, Mital A, Ramachandran U. Emerging targets for diabetes. Current science 2005; 88: 241-9.

34- Ruof J, Cieza A, Wolff B, Angst F, Ergeletzis D, Omar Z, et al. ICF Core Sets for diabetes mellitus . Journal of Rehabiliation Medicine 2004; 44: 100–6.

35- Motyl K, McCabe LR. Streptozotocin, Type I Diabetes Severity and Bone. Biological Procedures online 2009; 11: 296-315.

36- Tesch GH, Allen TJ. Rodent models of streptozotocin-induced diabetic nephropathy. Nephrology 2007: 12: 261-6.

37- Szkudelski T. The mechanism of alloxan and streptozotocin action in β-cells of the rat pancreas. Physiol Res. 2001; 50: 536-46.

38- Schroy PC, Cohen A, Winawer SJ, Friedman EA. New chemotherapeutic drug sensitivity assay for colon carcinomas in monolayer culture.Cancer Res 1988; 48: 3236– 44.

39- Dyke KV, Jabbour N, Hoeldtke R, Dyke CV, Dyke MV. Oxidative/nitrosative stresses trigger type I diabetes: Preventable in streptozotocin rats and detectable in human disease. Ann N Y Acad Sci 2010; 1203: 138–45.

40- Ledoux SP, Woodley SE, Patton NJ, Wilson GL. Mechanisms of nitrosourea-induced beta-cell damage. Alterations in DNA. Diabetes 1986; 35(8): 866–72.

41- Capucci MS, Hoffmann ME, Natarajan AT. Streptozotocin-induced genotoxic effects in Chinese hamster cells: The resistant phenotype of V79 cells. Mutation Research1995; 347: 79–85.

42- Ridolfi R, Amaducci L, Derni S, Fabbri L, Innocenti MP, Vignutelli P.Chemotherapy with 5-fluorouracil and streptozotocin in carcinoid tumors of gastrointestinal origin: Experience with 13 patients. Journal of Chemotherapy 1991; 3(5): 328–31.

43- Islam MS, Loots dT. Experimental rodent models of type 2 diabetes: A review. Methods Find Exp. Clin Pharmacol 2009; 31: 249–61.

44- Wei L, Lu Y, He S, Jin X, Zeng L, Zhang S, et al. Induction of diabetes with signs of autoimmunity in primates by the injection of multiple-low-dose streptozotocin. Biochem Biophys Res Commun 2011; 412(2): 373–8.

45- Hosokawa M, Dolci W, Thorens B. Differential sensitivity of GLUT1 and GLUT2-expressing beta cells to streptozotocin. Biochem Biophys Res Commun 2001; 289(5): 1114–7.

46- Friederich M, Hansell P, Palm F. Diabetes, oxidative stress, nitric oxide and mitochondria function. Curr Diabetes Rev 2009; 5: 120–44.

47- Simmons RA. Developmental origins of diabetes: The role of oxidative stress. Free Radic Biol Med 2006; 40: 917–22.

48- Raza H, Prabu SK, Robin MA, Avadhani NG. Elevated mitochondrial cytochrome P450 2E1 and Glutathione S-transferase A4-4 in streptozotocin-induced diabetic rats: Tissue-specific variations and role in oxidative stress. Diabetes 2004; 53: 185–94.

49- Raza H, Subbuswamy KP, John A, Avadhani NG. Impaired mitochondrial respiratory functions and oxidative stress in streptozotocin-induced diabetic rats. Int J Mol Sci 2011; 12: 3133–47.

50- Dyke KV, Ghareeb E, Dyke MV, Sosa A, Hoeldtke RD,Thiel DHV. Luminescence experiments involved in the mechanism of streptozotocin diabetes and cataract formation. Luminescence 2008; 23: 386–91.

51- Liu HK, McCluskey JT, McClenghan NH, Flatt PR. Streptozotocin-resistant BRIN-BD11 cells possess wide spectrum of toxin tolerance and enhanced insulin-secretory capacity. Endocrine 2007; 32: 20–9.

52- Frode TS, Medeiros YS. Animal models to test drugs with potential antidiabetic activity. Journal of Ethnopharmacology 2008; 115: 173–83.

53- Etuk EU. Animals models for studying diabetes mellitus. Agriculture and Biology Journal of North America 2010; 1: 130-4.

54- Grover JK, Vati V, Rathi SS, Dawar R. Traditional Indian antidiabetic plants attenuate renal hypertrophy,urine volume and albuminuria in streptozotocin-induced diabetic mice. J Ethnopharmacol 2001; 76: 233-8.

55- Wolff SP, Jiang ZY, Hunt JV. Protein glycation and oxidative stress in diabetes mellitus and ageing. Free Radic Biol Med 1991; 10:339-52.

56- Bloomer RJ, Goldfarb AH. Anaerobic exercise and oxidative stress: A review. Can J Appl Physiol 2004; 29(3): 245-63.

57- Baynes JW. Role of oxidative stress in development of complications in diabetes. Diabetes 1992; 40: 405-12.

58- Pari L, Saravanan R. Beneficial effect of succinic acid monoethyl ester on erythrocyte membrane bound enzymes and antioxidant status in streptozotocin-nicotinamide induced type 2 diabetes. Chem Biol Interact 2007;169:15-24

59 - Skrha J, Hodinar A, Krosnicka J, Hilgestora J. Relationship of Oxidative Stress and Fibrinolysis in Diabetes Mellitus. Diabet Med 1996; 13: 800-5.

60- Anderson D. Antioxidant defenses against reactive oxygen species causing genetic and other damage. Mutat Res 1996; 350:103-8.

61- Wohaieb SA, Godin DV. Alterations in free radical tissue defense mechanism in Streptozotocin -induced diabetes in rats. Effects of insulin treatment. Diabetes 1987; 36: 1014-8.

62- Halliwell B, Gutteridge JMC. Free radicals in biology and medicine, 4 th ed. Clarendron Press, Oxford 2006, pp. 1-541.

63- Haley S, Perret J, Harris M, Wilson J, Qian M. Free radical scavenging properties of wheat extracts. J Agric Food Chem 2002;50:1619-24.

64- Moussa S. Oxidative stress in diabetes mellitus. Romanian J biophys 2008; 18:225-36

65 - Nowier SR, Kashmiry NK, Abdel Rasool HA, Morad H, Ismail S. Association of type 2 diabetes mellitus and glutathione s transferase (GSTM1 and GSTT1) genetic polymorphism. Research Journal of Medicine and Medical Sciences 2009; 4: 181-8.

66- Gayatri M, kannabiran K. Anti-diabetic and Ameliorative Potential of Ficus bengalensis Bark extract in Streptozotocin- induced diabetic rats. IJCB 2008;23(4):394-400.

67- Karaca T, Yoruk M, Yoruk IH, Uslu S. Effects of green tea and ginseng on pancreatic beta cells and levels of serum glucose, insulin, cholesterol and triglycerides in rats with experimentally streptozotocin-induced diabetes: A histochemical and immunohistochemical study. J Animal and Veterinary Advances 2010; 9: 102-7.

68- Liu ZQ, Luo XY, Liu GZ, Chen YP, Wang ZC, Sun YX. In vitro study of the relationship between the structure of ginsenoside and its antioxidative or prooxidative

activity in free radical induced hemolysis of human erythrocytes. J Agric Food Chem 2003; 51:2555-8.

69- Cho WCS, Chung WS, Lee SKW. Ginsenoside Re of Panax ginseng possesses significant antioxidant and antihyperlipidemic efficacies in streptozotocin-induced diabetic rats. Eur J Pharmacol 2006; 550: 173-9.

70- DerMarderosian A. The Review of Natural Products by Facts and Comparisons. St. Louis, MO: Wolters Kluwer Co 1999, pp. 1-3

71- Lakshmi T, Anitha ROY, Geetha RV. *Panax ginseng* a universal panacea in the herbal medicine with diverse pharmacological spectrum – a review. Asian J Pharm Clin Res 2011;14: 14-8.

72- Jellin JM, Batz F, Hitchens K. Natural medicines comprehensive database 3rd Edition. Stockton, CA: Therapeutic Research Faculty 2002, pp. 1530.

73- Radad K, Gille G, Liu L, Rausch W. Use of ginseng in medicine with emphasis on neurodegenerative disorders. J Pharmacol Sci 2006; 100:175-86.

74- Yun TK. Brief introduction of *Panax ginseng* C.A. Meyer. Journal of Korean Medical Science supplement 2001;16: 3–5.

75- Park JD, Rhee DK, Lee YH. Biological activities and chemistry of saponins from *Panax ginseng C. A. Meyer*. Phytochemistry Reviews 2005; 4:159–75.

76- Boon H, Smith M. The Complete Natural Medicine Guide to the 50 Most Common Medicinal Herbs 2 nd ed. Toronto: Robert Rose 2004, pp.182-4.

77- Blumenthal M. Asian ginseng: potential therapeutic uses. Adv Nurse Pract 2001; 9(2):26-33.

78- Coon JT, Ernst E. *Panax ginseng*: a systematic review of adverse effects and drug interactions. Drug Saf 2002; 25: 323-44.

79- Vogler BK, Pittler MH, Ernst E. The efficacy of ginseng. A systematic review of randomized clinical trials. Eur J Clin Pharmacol 1999; 55:567-75.

80- Bahrke MS, Morgan WR. Evaluation of the ergogenic properties of ginseng: an update. Sports Med 2000; 29:113-33.

81- Ernst E. *Panax ginseng*: an overview of the clinical evidence. Journal of Ginseng Research 2010; 34(4): 259–63.

82- Choi KT. Botanical characteristics, pharmacological effects and medicinal components of Korean *Panax ginseng C A Meyer*. Acta Pharmacologica Sinica 2008;29(9):1109–18.

83- Lu JM, Yao Q, Chen C. Ginseng compounds: an update on their molecular mechanisms and medical applications.Curr Vasc Pharmacol 2009; 7: 293-302.

84- Dong-Hyun Kim. Chemical Diversity of *Panax ginseng*, *Panax quinquifolium*, and *Panax Notoginseng*. J Ginseng Res 2012;36: 1-15.

85- Blumenthal M. The ABC Clinical Guide to Herbs. New York, NY:Theime 2003, pp.11-25.

86- Lee TK, Johnke RM, Allison RR, Brien KFO, Dobbs LJ. Radioprotective potential of ginseng. Mutagenesis 2005; 20(4):237-43.

87- Duke JA. The Green Pharmacy Herbal Handbook: Your Comprehensive Reference to the Best Herbs for Healing. Emmaus PA: Rodale 2000, pp.115-6.

88- Shibata S. Chemistry and cancer preventing activitiesof ginseng saponins and some related, triterpenoidcompounds. J Korean Med Sci 2001;16:28-37.

89- WHO.*Panax ginseng* monograph. Alternative medicine Review 2009; 14(2): 172-6.

90- Chaudhary G, Goyal S, Poonia P. Lawsonia inermis Linnaeus: A Phytopharmacological Review. International Journal of Pharmaceutical Sciences and Drug Research 2010; 2(2): 91-8.

91- WHO . "Radix Ginseng" inWHO Monographs on Selected Medicinal Plants, Geneva. World Health Organization 1999; 1:168-82

92- Seely D, Dugoua JJ, Perri D, Mills E, Koren G. Safety and efficacyof *Panax ginseng* during pregnancy and lactation. CanJ Clin Pharmacol 2008; 15:87-94.

93- Kiefer D, Pantuso T. *Panax ginseng*. Am Fam Physician 2003; 68:1539-42.

94- Herrera DM, Abdala S, Benjumea D, Luis JG. Diuretic activity of some Withania aristata Ait.Fraction. Journal of Ethnopharmacology 2008; 117: 496-9.

95- Sotaniemi EA, Haapakoski E, Rautio A. Ginseng therapy in noninsulin- dependent diabetic patients. Diabetes Care 1995; 18:1373-5

96- Vuksan V, Sung MK, Sievenpiper JL, Jenkins AL, Buono MD, Lee KS, et al. Korean red ginseng (*Panax ginseng*) improves glucose and insulin regulation in well-controlled, type 2 diabetes: results of a randomized, Double-blind, placebo control ledstudy of efficacy and safety. Nutr Metab Cardiovasc Dis 2006; 18:46-56.

97- Hofseth LJ, Wargovich MJ. Inflammation, cancer, and targets of ginseng. J Nutr 2007; 137:183-5.

98- Keum YS, Han SS, Chun KS, Park KK, Park JH, Lee SK, et al. Inhibitory effects of the ginsenoside Rg3 on phorbol ester-inducedcyclooxygenase-2 expression, NF-kappaB activation and tumor promotion. Mutat Res 2003; 523-524:75-85.

99- Friedl R, Moeslinger T, Kopp B, Spieckermann PG. Stimulation of nitric oxide synthesis by the aqueous extract of *Panax ginseng* root in RAW 264.7 cells. Br J Pharmacol 2001; 134:1663-70.

100- Blumenthal M, Goldberg A, Brinkmann J. Herbal Medicine: Expanded Commission E Monographs. Austin, TX: Integrative Medicine Communications 2000, pp. 170-7.

101- Brinker FJ. Herb Contraindications and Drug Interactions. 3rd ed. Sandy, OR: Eclectic Medical Publications 2001, pp.107-9

102- Seely D, Dugoua JJ, Perri D, Mills E, Koren G. Safety and efficacy of *Panax ginseng* during pregnancy and lactation. Can J Clin Pharmacol 2008;15:87-94.

103- Mahady GB, Gyllenhall C, Fong HH, Farnsworth NR. Ginsengs: a review of safety and efficacy. Nutr Clin Care 2000;3:90-101.

104- Duke J. The Green Pharmacy Herbal Handbook: YourComprehensive Reference to the Best Herbs for Healing.Emmaus, PA: Rodale 2000, pp.115-6.

105- Omura T, Sato R. A new cytochrome in liver microsomes. J Biol Chem 1962; 237: 1375-6.

106- McKinnon RA, Sorich MJ, Ward MB. Cytochrome P450 Part 1: Multiplicity and Function. J Pharm Pract Res 2008; 38: 55-7.

107- Meunier B, devisser SP, Shaik, S. Mechanism of oxidation reactions catalyzed by cytochrome P450 enzymes.Chem Rev 2004;104: 3947-80.

108- Saxena A, Tripathi KP, Roy S, Khan F, Sharma A. Effects of herbal components on human drugs interactions involving Cytochrome P450. Bioinformation 2008; 3: 198-204.

109- Seliskar M, Rozman D. Mammalian cytochrome P450-importance of tissue specificity. Biochim Biophys Acta 2007; 3: 458-66.

110- Hannon-Fletcher MPA, O'Kane MJ, Moles KW, Barnett YA, Barnett CR. Lymphocyte cytochrome P450-CYP2E1 expression in human IDDM subjects. Food Chem Toxicol 2001; 39: 125-32.

111- Khan AJ, Sharma A, Choudhuri G, Parmar D. Induction of blood lymphocytes cytochrome P450 2E1 in early stage alcoholic liver cirrhosis. Alcohol 2011; 45: 81-7.

112- Murtomaa M, Viitala P, Hokkanen J, Pelkonen O, Rautio A. Xenobiotic metabolism of bank vole (Myodes glareolus) exposed to PCDDs. Environ Toxicol Pharmacol 2010; 29: 19-23.

113- Uppstad H, Øvrebø S, Haugen A, Mollerup S. Importance of CYP1A1 and CYP 1B1 in bioactivation of benzo[a]pyrene in human lung cell lines. Toxicol Lett 2010;192: 221-8.

114- Creshar B, Petric S. Cytochrome P450 enzymes in the fungal kingdom. Biochim Biophys Acta 2011; 1814: 29-35.

115- Guengerich FP, Zhongmei T, Qian C, Pinzon G diabetic. Approaches to deorphanization of human and microbial cytochrome P450 enzymes. Biochim Biophys Acta 2011; 1814: 139-45.

116- Nelson DR. Progress in tracing the evolutionary paths of cytochrome P450. Biochim Biophys Acta 2011; 1814: 14-8.

117- Hasler JA, Estabrook R, Murray M, Pikuleva I, Waterman M, Capdevila J, et al. Human cytochromes P450. Mol Asp Med 1999; 20: 1-137.

118- Nebert DW, Dalton TP. The role of cytochrome P450 enzymes in endogenous signalling pathways and environmental carcinogenesis. Nat Rev Cancer 2006; 6: 947-60.

119- Stiborova M, Rupertova M, Frei E. Cytochrome P450 and peroxidise-mediated oxidation of anticancer alkaloid ellipticine dictates its antitumor efficiency. Biochim Biophys Acta 2011;1814: 175-85.

120- Rooney PH, Telfer MC, Mcfadyen MC, Melvin WT, Murray GI. The role of cytochrome P450 in cytotoxic bioactivation: future therapeutic directions. Curr Cancer Drug Targets 2004; 4: 257-65.

121- Gum SI, Jo SJ, Ahn SH. The potent protective effect of wild ginseng (*Panax ginseng C.A.Meyer*) against benzo[α]pyrene-induced toxicity through metabolic regulation of CYP1A1 and GSTs. Journal of Ethnopharmacology 2007 ;112:568-76.

122- Jefferson JA, Shankland SJ, Pichler. Proteinuria in diabetic kidney disease: A mechanistic viewpoint. Kidney Int 2008; 74: 22-36.

123- Piccoli P, Carrieri M, Padovano L, DiMare M, Bartolucci GB, Fracasso ME, et al . In vivo CYP2E1 phenotyping as a new potential biomarker of occupational and experimental exposure to benzene. Toxicol Lett 2010; 192: 29-33.

124- Prieto-Castello MJ, Cardona A, Marhuenda D, Roel JM, Corno A. Use of the CYP2E1 genotype and phenotype for the biological monitoring of occupational exposure to styrene. Toxicol Lett 2010; 192: 34-9.

125- Ahn T, Yun CH, Oh DB. Tissue-specific of ascorbic acid supplementation on the expression of cytochrome P450 2E1 and oxidative stress in streptozotocin-induced diabetic rats. Toxicol Let 2006; 166(1):27-36.

126- Lukman AO, Salihu MA, Gafar AA, Aremu IT, Ayodele OS. Effect of vitamin C on malonadialdehyde (MDA) in pregnant Nigerian women. Journal of Basic and Applied Sciences 2008;4:105-8.

127- Qazi N, Neeru B, Imran M, Sanchit W,Harman K, Sheikh I. Malondialdehyde and superoxide dismutase levels-distinguishing parameters between benign and malignant pleural effusions. Free Radicals and Antioxidants 2012; 2:8-11.

128- Nnodin J, Anyadoh SO, Nwosu N.The antioxidant status and lipid peroxidation product of newly diagnosed and 6 weeks follow-up patients with pulmonary tuberculosis in Owerri, Imo State, Nigeria. Asian Pacific Journal of Tropical diseases 2011; 1 : 292 - 4.

129- Slatter D, Bolton CH, Bailey AJ. The importance of lipid-derived malondialdehyde in diabetes mellitus. Diabetologia 2000; 43:550–7.

130- Jain SK. In vivo externalization of phosphatidylserine and phosphatidyl thanolamine in the membrane bilayer and hypercoagulability by the lipid peroxidation of erythrocytes in rats. J Clin Invest1985; 76: 281–6.

131- Romero MJ, Bosch-Morell F, Romero B, Rodrigo J M, Serra MA, Romero FJ. Serum malondialdehyde: possible use for the clinical management of chronic hepatitis C patients. Free Radic Biol Med 1998; 25: 993–7.

132- Muchova J, Sustrova M, Garaiova I, Liptakova A, Blazicek P, Kvasnicka P, et al. Influence of age on activities of antioxidant enzymes and lipid peroxidation products in erythrocytes and neutrophils of Down syndrome patients. Free Radic Biol Med 2001; 31: 499– 508.

133- Marnett LJ. Oxy radicals, lipid peroxidation and DNA damage. Toxicology 2002; 181–182:219–22.

134- Tuma DJ. Role of malondialdehyde acetaldehyde adducts in liver injury. Free Radic Biol Med 2002; 32: 303–8.

135- Dib M, Garrel C, Favier A, Robin V, Desnuelle C. Can malondialdehyde be used as a biological marker of progression in neurodegenerative disease?. J Neurol 2002; 249:367–74.

136- Catherwood MA, Powell LA, Anderson P, Mcmaster D, Sharpe PC, Trimble ER. Glucose- induced oxidative stress in mesangial cells. Kidney Int 2002; 61: 599–608.

137- Jain SK, Morshed KM, Kannan K, Mcmartin KE, Bocchini JA, Effect of elevated glucose concentrations on cellular lipid peroxidation and growth of cultured human kidney proximal tubule cells. Mol Cell Biochem 1996; 162: 11–6.

138- Sharpe PC, Yue KK, Catherwood MA, Mcmaster D, Trimble ER. The effects of glucose- induced oxidative stress on growth and extracellular matrix gene expression of vascular smooth muscle cells. Diabetologia 1998;41:1210–9.

139- Hamilton JS, Powell LA, Mcmaster C, Mcmaster D, Trimble ER. Interaction of glucose and long chain fatty acids (C18) on antioxidant defences and free radical damage in porcine vascular smooth muscle cells in vitro. Diabetologia 2003; 46: 106–14.

140- Jain SK. Hyperglycemia can cause membrane lipid peroxidation and osmotic fragility in human red blood cells. J BiolChem 1989; 264: 21340–5.

141- Rajeswari P, Natarajan R, Nadler JL, Kumar D, Kalra VK. Glucose induces lipid peroxidation and inactivation of membrane-associated ion-transport enzymes in human erythrocytes in vivo and in vitro. J Cell Physiol1991; 149: 100–9.

142- Mohanty P, Hamouda W, Garg R, Aljada A, Ghanim H, Dandona P. Glucose challenge stimulates reactive oxygen species generation by leucocytes. J Clin Endocrinol Metab 2000; 85: 2970–3.

143- Dam PS, Asbeck BS, Oirschot JF, Biessels GJ, Hamers FP, Marx JJ. Glutathione and alpha-lipoate in diabetic rats: nerve function, blood flow and oxidative state. Eur J Clin Invest 2001; 31: 417–24.

144- Jachec W, Tomasik A, Tarnawski R, Chwalinska E. Evidence of oxidative stress in the renal cortex of diabetic rats:favourable effect of vitamin E. Scand J Clin Lab Invest 2002; 62: 81–8.

145- Karasu C. Increased activity of H_2O_2 in aorta isolated from chronically streptozotocindiabetic rats: effects of antioxidant enzymes and enzymes inhibitors. Free Radic Biol Med 1999; 27: 16–27.

146- Martin-Gallan P, Carrascosa A, Gussinye M, Dominguez C. Biomarkers of diabetes-associated oxidative stress and antioxidant status in young diabetic patients with or without subclinical complications. Free Radic Biol Med 2003; 34: 1563–74.

147- Wittenstein B, Klein M, Finckh B, Ullrich K, Kohlschutter A. Plasma antioxidants in pediatric patients with glycogen storage disease, diabetes mellitus, and hypercholesterolemia. Free Radic Biol Med 2002; 33: 103–10.

148- Sailaja YR, Baskar R, Saralakumari D. The antioxidant status during maturation of reticulocytes to erythrocytes in type 2 diabetics. Free Radic Biol Med 2003; 35:133–9

149- Sundaram RK, Bhaskar A, Vijayalingam S, Viswanathan M, Mohan R, Shanmugasundaram KR. Antioxidant status and lipid peroxidation in type II diabetes mellitus with and without complications. Clin Sci (Lond) 1996; 90: 255–60.

150- Olusi SO. Obesity is an independent risk factor for plasma lipid peroxidation and depletion of erythrocyte cytoprotectic enzymes in humans. Int J Obes Relat Metab Disord 2002; 26:1159–64.

151- Jain SK, Mcvie R. Hyperketonemia can increase lipid peroxidation and lower glutathione levels in human erythrocytes in vitro and in type 1 diabetic patients. Diabetes 1999; 48:1850–5.

152- Jain SK, Mcvie R, Jackson R, Levine SN, Lim G. Effect of hyperketonemia on plasma lipid peroxidation levels in diabetic patients. Diabetes Care 1999; 22:1171–5.

153- Jain SK, Kannan K, Lim G. Ketosis (acetoacetate) can generate oxygen radicals and cause increased lipid peroxidation and growth inhibition in human endothelial cells. Free Radic Biol Med 1998; 25: 1083–8.

154- Lima MH. Oxygen in Biology and Biochemistry: Role of Free Radicals. In: Storey KB, editor. Functional Metabolism: Regulation and Adaptation. New Jersey: Wiley–Liss, Inc. Hoboken 2004, pp. 319-68.

155- Haddad JJ. Oxygen sensing mechanism and the regulation of redox-responsive transcription factors in development and pathophysiology. Respir Res 2002; 3: 26-53.

156- Smith C , Marks A, Lieberman M. Basic Medical Biochemistry. A Clinical Approach. 2nd ed. Maryland: Lippincott Williams & Wilkins 2005, pp. 842-61.

157- Halliwell B, Gutteridge JMC. Oxygen is a Toxic Gas: An Introduction to Oxygen Toxicity and Reactive Species. In: Halliwell B & Gutteridge JMC editor. Free Radicals in Biology and Medicine. 4th ed. London: Oxford University Press 2007, pp. 21-2.

158- Lu SC.Regulation of glutathione synthesis. Molecular Aspects of Medicine 2009; 30: 42–59.

159- Oppenheimer L, Wellner VP, Griffith OW, Meister A. Glutathione synthetase. Purification from rat kidney and mapping of the substrate binding sites. The Journal of Biological Chemistry 1979; 254:5184–90.

160- Lu SC. Regulation of hepatic glutathione synthesis: current concepts and controversies. The FASEB Journal 1999; 13(10):1169–83.

161- Meister A. On the discovery of glutathione. Trends in Biochemical Sciences1988;13:185–8.

162- Townsend DM, Tew KD, Tapiero H. The importance of glutathione in human Disease. Biomedicine and Pharmacotherapy 2003;57 : 145–55.

163- Morel Y, Barouki R. Repression of gene expression by oxidative stress. Bio chemical Journal 1999; 342: 481–96.

164- Sen CK, Packer L. Antioxidant and redox regulation of gene transcription. The FASEB Journal 1996;10 (7) : 709–20.

165- Nikitovic D, Holmgren A, Spyrou G. Inhibition of AP-1 DNA binding by nitric oxide involving conserved cysteineresidues in Jun and Fos. Biochemical and Biophysical Research Communications 1998; 242(1): 109–12.

166- Rainwater R D, Parks M E, Anderson P, Tegtmeyer, Mann K. Role of cysteine residues in regulation of p53 function. Molecular and Cellular Biology 1995;15: 3892–903.

167- Wu HH, Momand J. Pyrrolidine dithiocarbamate prevents p53 activation and promotes p53 cysteine residue oxidation.The Journal of Biological Chemistry 1998;273(30) :18898–905.

168- Meister A, Anderson ME. Glutathione. Annual Review of Biochemistry 1983; 52 : 711–60.

169- Meister A.Glutathione deficiency produced by inhibition of its synthesis, and its reversal; applications in research and therapy. Pharmacology andTherapeutics 1991;51 :155–94.

170- Brigelius-Flohe R, Maiorino M. Glutathione peroxidases. Biochim Biophys Acta 2013;1830(5):3289-303.

171- Brigelius-Flohe R, Aumann KD, Blocker H, Gross G, Kiess M, Kloppel KD, et al. Phospholipid-hydroperoxide Glutathione peroxidase. Genomic DNA, cDNA, and deduced amino acid sequence. J Biol Chem 1994; 269(10):7342–8.

172- Ducros V, Favier A. Selenium metabolism. EMC Endocrinol Nutr 2004; 1: 19–28.

173- Meschy F. Nutrition minérale des ruminants; Editions Quae: Versaille, France 2010, pp. 208.

174- Flohe L. The Selenoprotein Glutathione Peroxidise. In Glutathione: Chemical, Biochemical and Medical Aspects, Part A; Dolphin D, Poulson R, Avramovic O, Eds; John Wiley & Sons Inc: New York NY, USA 1989, pp. 643–731.

175- Fairweather-Tait SJ, Collings R, Hurst R. Selenium bioavailability: Current knowledge and future research requirements. Am J Clin Nutr 2010; 91: 1484–91.

176- Bareither ML, Verhage HG. Control of the secretory cell cycle in cat oviduct by estradiol and progesterone. Am J Anat 1981; 162: 107–18.

177- Chu FF, Doroshow JH, Esworthy RS. Expression, characterization, and tissue distribution of a new cellular selenium-dependent glutathione peroxidase, GSHPx-GI. J Biol Chem 1993; 268: 2571–6.

178- Schwaab V, Faure J, Dufaure JP, Drevet JR. GPx-3: The plasma-type glutathione peroxidase is expressed under androgenic control in the mouse epididymis and vas deferens. Mol Reprod Dev 1998; 5: 362–72.

179- Maiorino M, Scapin M, Ursini F, Biasolo M, Bosello V, Flohe L. Distinct promotersdetermine alternative transcription of gpx-4 into phospholipid-hydroperoxide glutathione peroxidase variants. J Biol Chem 2003; 278: 34286–90.

180- Papp LV, Holmgren A, Khanna KK. Selenium and selenoproteins in health and disease. Antioxid Redox Signal 2010; 12: 793–5.

181- Ojiako OA, Nwanjo HU. Is Vernonia amygdalina hepatotoxic or protective? Response from biochemical and toxicity studies in rats. Afr J Biotechnol 2006; 5(18): 1648- 51.

182- Vozarova B, Stefan N, Lindsay SR, Saremi A, Pratley ER, Bogardus C, et al . High alanine aminotransferase is associated with decreased hepatic insulin sensitivity and predicts the development of type 2 diabetes. Diabetes 2002;51:1889-95.

183- Giboney PT. Mildly elevated liver transaminase levels in the asymptomatic patient. Am Fam Physician 2005; 71(6): 1105-10.

184- Satyanarayana U, Chakrapani U. Biochemistry. 3rd ed. Kolkatta (India): Books and Allied (P) Ltd; 2007, pp. 270-9.

185- Shirwaikar A, Rajendran K, Kumar CD, Bodla R. Antidiabetic activity of aqueous leaf extract of Annona squamosa in streptozotocin-nicotinamide type 2 diabetic rats. J Ethnopharmacol 2004; 91:171-5.

186- Pushparaj PN, Low HK, Manikandan J, Tan BK, Tan CH. Anti-diabetic effects of Cichorium intybus in streptozotocin-induced diabetic rats. J Ethnopharmacol 2007; 111: 430-4.

187- Karunanayake EH, Hearse DJ, Mellows G. The metabolic fate and elimination of streptozocin. Biochemical Society Transactions 1975; 3: 410-4.

188 - Yang CY, Wang J, Zhao Y, Shen L, Jiang X, Xie ZG, et al. Anti-diabetic effects of panax notoginseng saponions and its major antihyperglycemic components. J Ethnopharmacol 2010; 130: 231-6.

189- Hamilton K, Eaton EJ, Garland HO, Old S.Effects of experimental diabetes mellitus on Gentamicin-induced acute renal functional changes in the anesthetized rats.Clinical experimental Pharmacology and Physiology 1998; 25: 231–5.

190- Trinder P. Determination of blood glucose using an oxidaseperoxidase system with a non-carcinogenic chromogen. Indian J clin Path 1969; 22: 158-61.

191- Burtis CA, Ashwood ER, Bruns DE. Tietz Text Book of Clinical Chemistry and Molecular Diagnostics. 4th Ed. Elsevier Saunders Company. St Louis 2006, pp. 942-5.

192- Gella FJ, Olivella T, Cruz-Pastor M, Arenas J, Moreno R, Durban R, et al. A simple procerdure for routine determination of aspartate aminotransferase and alanine aminotransferase with pyridoxal phosphate. Clin Chem Acta 1985;153:241-7.

193- Lowry OH, Rosebrough NJ, Farr AL, Randall RJ. Protein estimation with the folin-ciocalteu reagent. J Biol CHEM 1951;193:448-50.

194- Sedlack J, Lindsay. Estimation of total protein bound and non-protein sulfhdryl groups in tissue with ellman s reagents. Anal Biochem 1968; 25:192-206.

195- Haaiwell B, Chirico S. Lipid peroxidation: its mechanism, measurement and significance. Am J Clin Nutr 1993; 57:7155-245.

196- Chiu DTY, Stults FH, Tappel AL. Purification and properties of rate lung soluble glutathione peroxidase. Biochem Biophys Acta 1976;445(3):558-66.

197- Haufroid V, Toubeau F, Clippe A , Buysschaert M, Gala J, Lison D. Real-time quantification of cytochrome P4502E1 mRNA in human peripheral blood lymphocytes by reverse transcription-PCR: Method and practical application. Clinical Chemistry 2001; 47: 1126–9.

198- Drury RAB, Wallington EA. Carleton 's histological technique, 5 th ed. Oxford university Press, Oxford New York Toronto 1980 , pp. 188-9,237-40, 290-1.

199- Leslie E, Geoffrey J, James M (eds). Statistical analysis. In: Interpretation and uses of medical statistics, 4th ed. Oxford Scientific Publications (pub) 1991, pp.411-6.

200- Kirkpatrick LA, Feeney BC. A simple guide to IBM SPSS statistics for version 20.0. Student ed. Belmont, Calif.: Wadsworth, Cengage Learning 2013, pp.115.

201- Barcelo A, Rajpathak S. Incidence and prevalence of diabetes mellitus in the Americas. Am J Public Health 2001; 10:300-8.

202- Weidmann P, Boehlen LM, De-courten M. Pathogenesis and treatment of hypertension associated with diabetes mellitus. Am Heart J 1993; 125:1498-513.

203- Kaneko H, Nakanishi K. Proof of the mysterious efficacy of ginseng: basic and clinical trials: clinical effects of medical ginseng, Korean red ginseng: specifically, its -anti stress actionfor prevention of disease. J Pharmacol Sci 2004; 95: 158-62.

204- Ohnishi Y, Takagi S, Miura T, Usami M, Kako M, Ishihara E, et al . Effect of ginseng radix on GLUT2 protein content in mouse liver in normal and epinephrine induced hyperglycemic mice. Biological & Pharmaceutical Bulletin 1996; 19: 1238- 40.

205- Xie JT, Wang CZ, Kim S, Yuan CS. The anti-hyperglycemic property of different ginseng partitions.Oriental pharmacy& Experimental Medicine 2005; 5: 1-15.

206- Woodcroft KJ, Hafner MS, Novak RF.Insulin signaling in the transcriptional and post-transcriptional regulation of CYP2E1 expression. Hepatology 2002; 35: 263–73.

207- Raza H, Ahmed I, John A, Sharma AK. Modulation of xenobiotic metabolism and oxidative stress in chronic streptozotocin-induced diabetic rats fed with Momordica charantia fruit extract. J Biochem Mol Toxicol 2000; 14: 131–9.

208- Ohkuwa T, Sato Y, Naoi M. Hydroxyl radical formation in diabetic rats induced by streptozotocin. Life Sci 1995; 56: 1789 –98.

209- Hodges NJ, Green RM, Chipman JK, Graham M. Induction of DNA strand breaks and oxidative stress in HeLa cells by ethanol is dependent on CYP2E1 expression. Mutagenesis 2007; 22: 189–94.

210- McKillop IH, Schrum LW. Alcohol and liver cancer . Alcohol 2005; 35: 195–203.

211- Stickel F, Schuppan D, Hahn EG, Seitz HK. Cocarcinogenic effects of alcohol in hepatocarcinogenesis.Gut 2002; 51: 132–9.

212- Seitz HK, Matsuzaki S, Yokoyama A, Homann N, Vakeva inen S, Wang XD. Alcohol and cancer. Alcohol Clin Exp Res 2001; 25: 137–43.

213- Ekstrom G, Ingelman-Sundberg M. Rat liver microsomal NADPH-supported oxidase activity and lipid peroxidation dependent on ethanol-inducible cytochrome P450. Biochem Pharmacol 1989; 38: 1313–9.

214- Szkudelski T. The mechanism of alloxan and streptozotocin action in B cells of the rat pancreas. Physiol Res 2001; 50:537-46.

215- Maiti R, Jana D, Das UK, Ghosh D. Antidiabetic effect of aqueous extract of seed of Tamarindus indica in streptozotocin induced diabetic rats. J Ethnopharmacol 2004; 92: 85-91.

216- Aughsteen AA. An ultrastructural study on the effect of STZ on the islet of Langerhans in mice. Electric Microscope 2000; 49: 681-90.

217- Attia AA. Histological and electron microscopic studies of the effect of B-Carotene on the pancreas of streptozotocin (STZ)-induced diabetic rats. Pak J Biol Sci 2009; 12: 301-14.

218- Yoon M, Lee H, Jeong S, Kim JJ, Nicol CJ, Nam KW, et al. Peroxisome proliferator-activated receptor alpha is involved in the regulation of lipid metabolism by ginseng. Br J Pharmacol 2003; 138: 1295-302.

219- Kintscher U, Law RE. PPARgamma-mediated insulin sensitization: the importance of fat versus muscle. Am J Physiol Endocrinol Metab 2005; 288: 287-91.

220- Lee H, Gonzalez FJ, Yoon M. Ginsenoside Rf, a component of ginseng, regulates lipoprotein metabolism through peroxisome proliferator-activated receptor alpha. Biochem Biophys Res Commun 2006; 339: 196-203.

221- Park MY, Lee KS, Sung MK. Effects of dietary mulberry, Korean red ginseng, and banaba on glucose homeostasis in relation to PPAR-α, PPAR-γ, and LPL mRNA expressions. Life Science 2005; 77:3344–54.

222- Han KL, Jung MH, Sohn JH, Hwang JK. Ginsenoside 20S-protopanaxatriol (PPT) activates peroxisome proliferator-activated receptor gamma (PPARgamma) in 3T3-L1 adipocytes. Biol Pharm Bull 2006; 29: 110-3.

223- Li AC, Glass CK. PPAR- and LXR-dependent pathways controlling lipid metabolism and the development of atherosclerosis. J Lipid Res 2004; 45: 2161-73.

224- Michalik L, Auwerx J, Berger JP, Chatterjee VK, Glass CK, Gonzalez FJ, et al. International union of pharmacology. LXI. Peroxisome proliferator-activated receptors. Pharmacol Rev 2006; 58: 726-41.

225- El-Khayat Z, Hussein J, Ramzy T, Ashour M, Oraby F. Protective Effect of *Panax Ginseng* Against Streptozotocin Induced Renal Dysfunction in Rats. J Appl Sci Res 2011;7: 1419-23.

226- Ye J. Role of insulin in the pathogenesis of free fatty acid- induced insulin resistance in skeletal muscle. Endocrine, Metabolism & Immune disorders Drug Targets 2007; 7: 65-74.

227- Zhang Z, Li X, Lv W, Yang Y, Gao H, Yang J, et al. Ginsenoside Re reduces insulin resistance through inhibition of JNK and NF-KB. Mol Endocrinol 2008; 22: 186–95.

228- Davydov VV, Molokovski DS, Limarenko A. Effeicacy of ginseng drug in experimental insulin dependant diabetes and toxic hepatitis. Pathologicheskaia Fiziologiia i EksperimentalnaiaTerapiia 1990; 5: 49-52.

229- Lee WK, Kao ST, Liu IM, Cheng JT. Increase of insulin secretion by insenoside Rh2 to lower plasma glucose in Wistar rats. Clinical &experimental Pharmacology & Physiology 2006; 33: 27-32.

230- Sathishsekar D, Subramanian S. Beneficial effects of Momordica charantia seeds in the treatment of STZ-induced diabetes in experimental rats. Biol Pharm Bull 2005; 28:978-83.

231- Yao HT, Huang SY, Chiang MT. A comparative study on hypoglycemic and hypocholesterolemic effectsof high and low molecular weight chitosan in streptozotocin-induced diabetic rats. Food Chem Toxicol 2008 ;46:1525-34.

232- Kim SH, Kang JS, Lee SJ, Chung YJ. Antidiabetic effect of Korean red ginseng by puffing process in streptozotocin-induced diabetic rats. J Korean Soc Food Sci Nutr 2008; 37:701-7.

233- Lemieux J, Pichette C, Vinay P, Gougoux A. Cellular Mechanism of antiammoniagenic effect of ketone bodies in the dog. Am J PhysiolRenal 1980; 239:420-6.

234- Latha S, Rajaram K, Suresh KP.Hepatoprotective and antidiabetic effect of methanol extract of Caralluma Fimbriata in streptozotcin induced diabetic albino rats. J Pharm Pharm Sci 2014; 1: 665-8.

235- Maritim AC, Sanders RA, Watkins JB. Diabetes, oxidative stress, and antioxidants: a review. J Biochem Toxicol 2003; 17: 24-38.

236- Jung CH, Zhou S, Ding GX, Kim JH, Hong MH Shin YC, et al. Antihyperglycemic activity of herb extracts on streptozotocin-induced diabetic rats. Biotechnol Biochem 2006; 70: 2556-9.

237- Garella S. The cost of dialysis in the USA. Nephrol DialTransplant 1997; 12:10-2.

238- Das AV, Padayatti PS, Paulose CS. Effect of leaf extract of Aegle Marmelose (L.) Correaex Roxb. Onhistological and ultrastructural changes in tissues of Streptozotocin induced diabetic rats. Indian J Exp Biol 1996; 34(4):341-5.

239- Degirmenchi I, Kalender S, Ustuner MC, Kalender Y, Gunes HV, Unal N, Basaran A. The effects of acarbose and Rumex patientia on liver ultrastructure in streptozotocin-induced diabetic (type-II) rats. Drugs Exp Clin Res 2002; 28(6):229-34.

240- Kim HJ, Chae IG, Lee SG, Jeong HJ, Lee EJ, Lee IS. Effects of Fermented Red Ginseng Extracts on Hyperglycemia in Streptozotocin-induced Diabetic Rats. J Ginseng Res 2010; 34: 104-12.

241- Kwak YS, Kyung JS, Kim JS, Cho JY , Rhee MN . Anti-hyperlipidemic effects of red ginseng acidic polysaccharides from Korean red ginseng. Biol Pharmaceut Bull 2010; 33(3):468-72.

242- Kim HJ, Chae IG, Lee SG, Jeong HJ, Lee EJ, Lee IS. Antioxidant Effects of Fermented Red Ginseng Extracts in Streptozotocin- Induced Diabetic Rats. J Ginseng Res 2011 ; 35: 129-37.

243- Kim JH, Chun YJ, Park JD, Kim SI, Roh JK, Jeong TC. Protection of rat liver microsomes againstcarbon tetrachloride-induced lipid peroxidation by red ginseng saponins through cytochrome P450 inhibition. Planta Medica 1997; 63:415-8.

244- Lee AY, Chung SS. Contributions of polyol pathway to oxidative stress in diabetic cataract. FASEB J 1999; 13: 23-30.

245- Ceriello A, Quatraro A, Giugliano D. New insights on nonenzymatic glycosylation may prevention of diabetic complications. Diabet Med 1992; 9: 297-9.

246- Winterbourn CC. Concerted antioxidant activity of glutathione and superoxide dismutase. In: Packer, L, Fuchs, J. (Eds.) Biothiols in Health and Disease. Marcel Dekker Inc: New York, NY 1995, pp. 117-34.

247- Arthur JR. The glutathione peroxidases. Cell. Mol Life Sci 2000;57: 1825–35.

248- Jachec W, Tomasik A, Tarnawski R, Chwalinska E. Evidence of oxidative stress in the renal cortex of diabetic rats: favourable effect of vitamin E. Scand. J Clin Lab Invest 2002; 62: 81–8.

249- Obrosova IG, Fathallah L, Liu E, Nourooz-Zadeh J. Early oxidative stressin the diabetic kidney: effect of DL-alpha-lipoic acid. Free Radic Biol Med 2003; 34: 186–95.

250- Shirwaikar A, Rajendran K, Kumar D, Bodla R. Antidiabetic activity of aqueous leaf extract of Annona squamosa in dtreptozotocin-nicotineamide type 2 diabetic rats. J Ethnopharmacol 2004; 91:152-78.

251- Kim JS, Kim KW, Choi KJ, Kwak YK, Lee KH, Chung HY.Screening of antioxidative components from red ginseng saponin. Korean J Ginseng Sci 1996; 20: 173-8.

252- Zhang HA, Wang M, Zhou J, Yao QY, Ma JM, Jiang CL. Protective effect of ginsenoide against acute renal failure and expression of tyrosine hydroxylase in the locus coeruleus. Physiol Res 2010; 59:61-70.

253- Liu ZQ, Luo XY, Liu GZ, Chen YP, Wang ZC, Sun YX. In vitro study of the relationship between the structure of ginsenoside and its antioxidative or prooxidative activity in free radical induced hemolysis of human erythrocytes. J Agric Food Chem 2003; 51: 2555-8.

254- Surh YJ, Na HK, Lee JY, Keum YS. Molecular mechanisms underlying anti-tumor promoting activities of heat-processed Panax ginseng C.A Meyer. J Korean Med Sci 2001; 16: 38-41.

255- Raza H, Prabu SK, Robin MA, et al. Elevated mitochondrial Cytochrome P450 2E1 and Glutathione S-transferase A4-4 in Streptozotocin-induced diabetic rats. Diabetes 2004; 53: 185-93.

256- Wang Z, Hall SD, Maya JF,LI L, Asghar A, Gorski JC. Diabetes mellitus increases the in vivo activity of Cytochrome P450 2E1 in humans. Br J Clin. Pharmacol 2003; 55(1): 77-85.

257- Barnett CR, Petrides L, Wilson J, Flatt PR, Ioannides C. Induction of rat hepatic mixed-function oxidases by acetone and other physiological ketones: their role in diabetes-induced changes in cytochrome P450 proteins. Xenobiotica 1992; 22: 1441–50.

258- Shimojo N, Ishizaki T, Imaoka S, Funae Y, Fuji S, Okuda K. Changes in amounts of cytochrome P450 isozymes and levels of catalytic activities in hepatic and renal microsomes of rats with streptozocin-induced diabetes. Biochem Pharmacol 1993; 46: 621–7.

259- Enriquez A, Leclercq I, Farrell GC, Robertson G. Altered expression of hepatic CYP2E1 and CYP4A in obese, diabetic ob/ob mice and fa/fa Zucker rats. Biochem Biophys Res Commun 1999; 255: 300–6.

260- Barnett CR, Gibson GG, Wolf CR, Flatt PR, Ioannides C. Induction of cytochrome P450III and P450IV family proteins in streptozotocin-induced diabetes. Biochem J 1990 ; 268: 765– 9.

261- Shimojo N. Cytochrome P450 changes in rats with streptozocin-induced diabetes. Int J Biochem 1994; 26: 1261– 8.

262- Mari M, Cederbaum AI. Induction of catalase, alpha and microsomal glutathione S-transferase in CYP 2E1 overexpressing HepG2 cells and protection against short-term oxidative stress. Hepatology 2001; 33:652–61.

263- Sadi G, Sadi O. Reciprocal Modulation of Hepatic Cytochrome P450 Gene Expressions with Streptozotocin Induced Diabetes and Resveratrol. Journal of Applied Biological Sciences 2010; 4: 55-62.

264- Woodcroft JK, Hafner MS, Novak RF. Insulin signaling in the transcriptional and posttranscriptional regulation of CYP2E1 expression. Hepatology 2002;35: 263-73.

265- Wang YG, Gao Y, Chai BX, Chen P, Tan HL, Zhao YH . Modulation of the activity and mRNA expression of cytochrome P450 isozymes in rat liver by *Panax ginseng* and co-administration with Veratrumnigrum. ZhongguoZhongyaoZazhi 2004; 29: 366–70.

266- Shim JY, Kim MH, Kim HD, Ahn JY, Yun YS, Song JY. Protective action of the immunomodulator ginsan against carbon tetrachloride-induced liver injury via control of oxidative stress and the inflammatory response. Toxicology and Applied Pharmacology 2010; 242:318–25.

267- Kim HG, Yoo SR, Park HJ, Lee NH, Shin JW, Cho JH, et al. Antioxidant effects of *Panax ginseng C.A. Meyer* in healthy subjects: a randomized, placebo-controlled clinical trial. Food and Chemical Toxicology 2011; 49(9): 2229–35.

268- Liu Y, Zhang JW, Li W, Ma H, Sun J, Deng MC, et al. Ginsenoside metabolites, rather than naturally occurring ginsenosides, lead to inhibition of human cytochrome P450 enzymes. Toxicological Sciences 2006; 91(2) : 356–64.

269- Wang T, Shankar K, Ronis MJ, Mehendale HM. Mechanisms and outcomes of drug- and toxicant-induced liver toxicity in diabetes. Crit Rev Toxicol 2007; 37: 413–59.

270- Weltman MD, Farrell GC, Hall P, Ingelman-Sundberg M, Liddle C. Hepatic cytochrome P450 2E1 is increased in patients with nonalcoholic steatohepatitis. Hepatology 1998; 27: 128–33.

271- Niemela O, Parkkila S, Juvonen RO, Viitala K,Gelboin HV, Pasanen M. Cytochromes P450 2A6, 2E1 and 3A and production of protein-aldehyde adducts in the liver of patients with alcoholic and nonalcoholic liver diseases. J Hepatol 2000; 33: 893–901.

272- Leclercq IA, Farrell GC, Field J, Bell DR, Gonzalez FJ, Robertson GR. CYP2E1 and CYP4A as microsomal catalysts of lipid peroxides in murine nonalcoholic steatohepatitis. Journal of Clinical Investigation 2000; 105(8): 1067–75.

تأثير مستخلص الجنسنج علي سيتوكروم ب ٤٥٠ ٢ أى ١ وبعض الدلائل الحيوية للإجهاد التأكسدي في الفئران المصابة بمرض السكر المستحدث بالستربتوزوتسين

The effect of panex ginseng extract on Cytochrome P450 2E1 and some biomarkers of oxidative stress in streptozotocin-induced diabetic rats

Protocol of a thesis submitted to the	خطة بحث مقدمة إلى
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Background

Diabetes is a major degenerative disease in the world today, affecting at least 15 million people and having complications which include hypertension, atherosclerosis and microcirculatory disorders.[1] Diabetes mellitus is also associated with long-term complications, including retinopathy, nephropathy, neuropathy and angiopathy and several others .[2]

Panax ginseng C.A Mayer has been widely used in traditional oriental herbal medicine in many countries.[3] The pharmacological properties of ginseng are mainly attributed to ginsenosides, the active components found in the extracts of different species of ginseng.[4] The extracts of ginseng have been found to possess cardioprotective, antiasthmatic, antidiabetic and potent central nervous system activities including enhancement of memory, concentration, mental alertness and decrease of mental fatigue, besides other beneficial activities.[5] It is well known that the ginseng antioxidant properties are scavenging free radicals and neutralizing ferric ion-induced peroxidation. [6] These antioxidant actions contribute to prevention and treatment of diseases associated with oxidative stress.[7]

Liver is a focal organ in oxidative and detoxifying processes as well as free radicals reactions and the biomarkers of oxidative stress are elevated in the liver at an early stage in many diseases, including diabetes mellitus.[8]

Oxidative stress is involved in the etiology and pathogenesis of many diabetic complications.[9] Streptozotocin(STZ) injection in animals generates type1 diabetes and produces various kinds of reactive oxygen species (ROS) that are either formed by STZ itself over the short term or resulted from the induced hyperglycemia.[10] Type1 diabetes mellitus (T1DM) is an autoimmune disease that is characterized by specific destruction of ß-cells in the islets of Langerhans.[11]

The increased oxidative stress, as measured by indices of elevated lipid peroxidation products such as malondialdhyde (MDA) ,oxidative modification products of proteins, depletion of endogenous antioxidant, are commonly found in STZ-induced diabetic rats, and these alterations may cause tissues to be more susceptible to oxidative damage .The vulnerability of each tissue to oxidative stress can vary depending upon their expressed antioxidant enzymes such as glutathione peroxidase (GPx).[12]

The cytochrome P450 superfamily (officially abbreviated as CYP) is a large and diverse group of enzymes. The function of most CYP enzymes is to catalyze the oxidation of organic substances. The substrates of CYP enzymes include metabolic intermediates such as lipids and steroidal hormones, as well as xenobiotic substances such as drugs and other toxic chemicals. CYPs are the major enzymes involved in drug metabolism and bioactivation, accounting for ~75% of the total number of different metabolic reactions.[13] The Cytochrome P450 2E1 has been implicated in the generation of tissue damaging hydroxyl radicals in patients suffering from diabetes and liver diseases.[14]

Aim of the work

The aim of the present work is to investigate the effect of panex ginseng extract on Cytochrome P450 2E1 and some oxidative stress markers in streptozotocin-induced diabetic rats.

Materials and Methods

1 –Experimental animals:

Fifty healthy adult male albino rats approximately 3 months old, weighing (120 ± 5 gm), will be used in the present study. They will be kept in special plastic rodent cages in laboratory under nearly constant conditions for experimental work. Animals will be kept in laboratory for at least one week before initiation of the experiments and will be maintained on a standard rodent diet and clean water.

The study was approved by the ethical committee of Medical Research Institute .

The animals will be divided into five groups:

Group I : consisting of 10 rats and serve as normal controls .

Group II : consisting of 10 diabetic rats (diabetes will be induced by a single intraperitoneal injection of streptozotocin at a dose of 60 mg/kg body weight for 3 consecutive days).[15]

Group III : consisting of 10 rats that will receive panax ginseng extract dissolved in distilled water (a dministered orally at dose of 100 mg/kg body weight for 30 days).[16]

Group IV : consisting of 10 rats pretreated with panax ginseng extract as in group **III** for 30 days before their injecting by streptozotocin as in group **II** for 3 consecutive days.

Group V : consisting of 10 diabetic rats as in group **II** , that will be treated with a panax ginseng extract as in group **III** for 30 days **.**

* Body weights of rats will be recorded at the beginning and at the end of the experiment period (33 days) .
* It should be noted that rats in group 1, 2 will be orally administrate with distilled water in a volume matched with that given to panax ginseng extract groups.

2 – Biochemical study:

At the end of the experiment, the blood will be collected from the heart of each rat partly in plain tubes to obtain serum and partly on K3 EDTA coated tubes to obtain plasma, used for determination of :-

1- Fasting plasma glucose level.[17]

2- Total serum cholesterol and triglycerides levels.[18]

3- Serum ALT and AST activities.[19]

Liver tissues examination :

Liver tissues will be separated from rats and will be cut into four parts and used as follow :

* First part of liver will be homogenized in 10 mM potassium phosphate containing 1 mM EDTA, pH 7.4 and centrifuged at 12.000 xg for 30 min at 4 ^{O}C and used for determination of:-
  - Malondialdhyde (MDA) level.[20]
  - Glutathione peroxidase (GPx) enzyme activity.[21]
  - Glutathione (GSH) content.[22]

* Second part of the liver will be used for microsomes preparation, which will be used for determination of :-
  - Cytochrome P450 2E1 (CYP 2E1) enzyme activity.[23]

* Third part of the liver will be used for determination of:-
  - Cytochrome P450 2E1 (CYP 2E1) gene expression by reverse transcription PCR.[24]

3 - Histopathological analysis.

* Part of pancreas and fourth part of liver specimens will be extracted, placed in formalin solution, and processed routinely by embedding in paraffin. Tissue sections (4-5 μ m) will be stained with haematoxylin-eosin and examined under light microscope.[25]

Analysis of the results

The obtained data will be tabulated and statistically analyzed using SPSS software .

References

1- Ogbonnia, S O , Odimegwu, J I , Enwuru V N . Evaluation of hypoglycemic and hypolipidemic effects of ethanolic extracts of Treculia africana Decne and Bryophyllum pinnatum Lam. and their mixture on streptozotocin (STZ) - induced diabetic rats. African Journal of Biotechnology 2008; 7 : 2535 - 9.

2- Kristova V, Liskoya S,Sotnikova S,Vojtko R, KurtanskyA. Sulodexide improves Endothelial Dysfunction in Streptozotocin- Induced Diabetes in Rats. Physiol Res 2008; 5: 491-4.

3- Kim DH, Jung JS, Moon YS, Sung JH, Suh HW, Kim YH, et al . Inhibition of intracerebroventricular injection stress-induced plasma corticosterone levels by intracerebroventricularly administered compound K, a ginseng saponin metabolite, in mice. Biol Pharm Bull 2003; 26:1035–8.

4- Nah SY, Kim DH, Rhim H. Ginsenosides: Are Any of them Candidates for Drugs Acting on the Central Nervous System .CNS Drug Rev 2007; 13: 381–404.

5- Gillis CN. Panax ginseng pharmacology: A nitric oxide link. Biochem Pharmacol 1997; 54: 1-8.

6- Jeong TC , Kim H J, Park J, Suha C, Park JD, Kim S, et al . Protective effects of red ginseng saponins against carbon tetrachloride-induced hepatotoxicity in Sprague-Dawley rats .Planta Med 1997; 63: 136-40.

7- Mahady GB, Gyllenhall C, Fong HH , Farnsworth NR. Ginsengs : a review of safety and efficacy. Nutr Clin Care 2000; 3: 90-101.

8- Kume E, Fujimura H, Matsuki N, Ito M, Aruga C, Toriumi W, et al. Hepatic changes in acute phase of streptozotocin (SZ)-induced diabetes in mice . Exp Toxicol Pathol 2004; 55: 467-80.

9 - Niedowic DM, Daleke DL. The role of oxidative stress in diabetic complications. Cell Biochem Biophys 2005; 43:289-330.

10- Piconi L, Quagliara L,Ceriello A. Oxidative stress in diabetes . lin Chem Lab Med 2003 ; 41:1144-9.

11- Atkinson MA , Maclaren NK. The pathogenesis of insulin dependent diabetes mellitus. N Engl J Med 1994; 331:1428–36.

12- Damasceno DC ,Volapato GT, Paranhos-Caderon M, Cunha-Rudge MV. Oxidative stress and diabetes in pregnant rats. Anim Reprod Sci 2002; 72:235-44.

13- Guengerich FP . Cytochrome p450 and chemical toxicology. Chem Res Toxico 2008; 21 : 70– 83.

14- Ahn T, Yun CH, Oh DB . Tissue-specific effect of ascorbic acid supplementation on the expression of cytochrome P450 2E1 and oxidative stress in streptozotocin-induced diabetic rats.Toxicol Lett 2006;166 : 27-36.

15- Karunanayake EH, Hearse D J, Mellows G. The metabolic fate and elimination of streptozocin. Biochemical Society Transactions 1975; 3: 410-4.

16- Yang CY, Wang J, Zhao Y, Shen L, Jiang X, Xie ZG, et al. Anti-diabetic effects of panax notoginseng saponions and its major antihyperglycemic components . J Ethnopharmacol 2010;130: 231-6.

17 Bopanna KN , Kannan J , Gadgil S , Balaraman ER , Rathore SP . Antidiabetic and antihyperglycaemic effects of neem seeds kernel powder on alloxan diabetic rabbits. Indian Journal of Pharmacology 1997; 29: 62-7.

18- Burtis CA, Ashwood ER, Bruns DE. Tietz Text Book of Clinical Chemistry and Molecular Diagnostics. 4th Ed. Elsevier Saunders Company . St Louis 2006, pp. 942-5 .

19- Gella FJ , Olivella T , Cruz-Pastor M , Arenas J , Moreno R , Durban R, et al . A simple procerdure for routine determination of aspartate aminotransferase and alanine aminotransferase with pyridoxal phosphate. Clin Chem Acta 1985;153:241-7.

20- Haaiwell B , Chirico S.Lipid peroxidation: its mechanism, measurement and significance . Am J Clin Nutr 1993;57:7155-245.

21- Flohe L,Gunzler WA . Assays for glutathione peroxidase . Methods Enzymol 1984 ; 105 : 14-21.

22- Griffith OW. Determination of glutathione disulphide using glutathione reductase and 2-vinylpyridine. Analytical Biochemistry 1980;106: 207-12.

23- Reinke LA , Moyer MJ. p-Nitrophenol hydroxylation: a microsomal oxidation which is highly inducible by ethanol . Drug Metab Dispos 1985;13:548-52.

24- Yang SP, Ranger GM . Cytochrome P450 expression and activities in human tongue cells and their modulation by green tea extract . Toxicol Appl Pharmacol. 2005; 202 : 140-50.

25- Lillie RD, Fulmer H M. Histopathological Technique and Practical Histochemistry . 4th ed. McGraw Hill, New York 1976, pp. 205 – 8 .

الملخص العربي

يتميز مرض البول السكري بارتفاع معدل الجلوكوز في الدم نتيجة خلل في انتاج هرمون الانسولين أو تاثير هرمون الأنسولين أو كليهما والذي يؤدي الي مشاكل وعائية عظمي أو صغري ، فهو مرض مزمن يهدد ظروف الحياة و يعتمد علي الأدوية و النظام الغذائي و نظام الحياة ليمنع المضاعفات علي المدي البعيد .

لو لم يعود اختلال توازن الجلوكوز الي المعدل الطبيعي في مده من الوقت فذلك يؤدي الي ارتفاع نسبه السكر في الدم و يسمي مرض البول السكري.

يعتبر الجنسنج علاج عشبي شعبي مستخدم منذ الاف السنين . فهناك خمس أنواع رئيسيه من الجنسنج فمنه الأمريكي والصيني والكوري والياباني و الروسي .

وهذا العشب له قاعدة واسعة من التطبيقات والذي يعتبر الاكثر شيوعا في طب الأعشاب علي مستوي العالم ، فهو يستخدم في علاج كثيرا من الاضطرابات متضمنا الأنيميا والأرق وصعوبة التنفس وضعف الذاكرة و الأرتباك وانخفاض الرغبة الجنسية والتعب المزمن والذبحة الصدرية ومرض البول السكري .

يعتبر سيتوكروم ب ٤٥٠ ٢ أى ١ (CYP2E1) من عائلة سيتوكروم ب ٤٥٠ ، التي لها دورا رئيسيا في إزالة السموم والتنشيط الحيوي للادويه والمواد الغريبة.

الهدف من البحث :هو دراسة تاثير مستخلص الجنسنج علي التعبير الجيني لسيتوكروم ب ٤٥٠ ٢ أى ١ وبعض مؤشرات الإجهاد التأكسدي مثل (المالون داي ألدهيد) وبعض مضادات الأكسده مثل (الجلوتاثيون والجلوتاثيون بيروكسيديز) في الفئران المصابة بمرض السكر المستحدث بالستربتوزوتسين.

مواد وطرق البحث :

تمت هذه الدراسة علي عدد ٥٠ فأر وتم تقسيمهم الي ٥ مجموعات رئيسيه وهي كالاتي:

المجموعة الاولي: تتكون من ١٠ فئران وهي تمثل (المجموعه الضابطه).

المجموعة الثانيه : تتكون من ١٠ فئران مصابه بمرض البول السكري (تم حقنها داخل الغشاء البريتوني بماده الستربتوزوتسين ٦٠ مجم لكل كيلو جرام من وزن الفئران لمدة ٣ أيام متتاليه) (المجموعه المصابه بمرض البول السكري).

المجموعة الثالثه : تتكون من ١٠ فئران اعطيت مستخلص نبات الجنسج يوميا تركيزه ٤ % مذاب في ماء مقطر (اعطيت بالفم ١٠٠ مللي جرام لكل كيلو جرام من وزن الفئران لمدة ٣٠ يوما) (مجموعه الجنسنج).

المجموعة الرابعه : تتكون من ١٠ فئران تم علاجها أولا بمستخلص الجنسنج كما في المجموع الثالثه لمدة ٣٠ يوما قبل حقنها بماده الستربتوزوتسين كما في المجموعه الثانيه لمده ٣ أيام (المجموعه المعالجه اولا قبل الاصابه بمرض البول السكري).

المجموعة الخامسه: تتكون من ١٠ فئران تمت اصابتها بمرض البول السكري أولا كما في المجموعه الثانيه ثم عولجت بمستخلص الجنسنج كما في المجموعة الثالثه لمدة ٣٠ يوما (المجموعه المصابه بمرض البول السكري المعالجة بالجنسنج).

عند مقارنة الأوزان النهائية للفئران مع الأوزان الأولية لها وجد أن وزن الجسم قد زاد زياده ذو دلاله احصائية في المجموعة الضابطه ومجموعة الجنسنج والمجموعة المعالجه اولا بالجنسنج قبل الاصابة بمرض البول السكري وقد قل وزن الفئران في مجموعة المصابة بمرض البول السكري والمجموعه المعالجه بالجنسنج بعد الاصابة بمرض البول السكري .

وبمقارنة قيم الاوزان النهائية للمجموعات مع المجموعة الضابطه نقصت نقصا ملحوظا في المجموعه المصابة بمرض البول السكري وزادت زياده واضحة في مجموعة الجنسنج، ولكنها لم تبدي اي اختلاف واضح في كلا من

المجموعة الرابعه والخامسه. وبالمقارنة بالمجموعة المصابة بمرض البول السكري وجد ان وزن الجسم النهائي قد زاد زياده واضحة في مجموعة الجنسنج ولكن لم يبدو اي اختلاف واضح في المجموعة الرابعه والخامسه .

تم إجراء الدراسات التالية في كل مجموعات الدراسة:

الدراسات البيوكيميائية:

تم تعيين مستوي الجلوكوز الصائم و الكوليستيرول والدهون الثلاثية و نشاط انزيمات الألانين ترانسفيراز ALT والأسبارتات ترانسفيراز AST في مصل الفئران .

و الجلوتاثيون والجلوتاثيون بيروكسيديز و المالون داي ألدهيد و التعبير الجيني لسيتوكروم ب ٤٥٠ ٢ أى ١ في خلايا الكبد وقد تم فحص مجهري لنسيج الكبد والبنكرياس في كل المجموعات السابقه لدراسة التغيرات الهستوباثولوجية .

وقد أسفرت الدراسة علي النتائج التالية:

١- **في المجموعة الثانيه (المجموعة المصابه بمرض البول السكري)**: قيم كلا من مستوي الجلوكوز الصايم والكوليستيرول والدهون الثلاثية و المالون داي ألدهيد ونشاط ALT و AST و التعبير الجيني لسيتوكروم ب ٤٥٠ ٢ أى ١ زادت زيادة ملحوظة بالمقارنه بالمجموعه الضابطه بينما نقصت قيم كلا من الجلوتاثيون والجلوتاثيون بيروكسيديز .

٢- **في المجموعة الثالثة (مجموعه الجنسنج)**: لم تتغير قيم المعايير البيوكيميائيه عند مقارنتها بالمجموعه الضابطه بينما نقصت كل القيم نقصا ذو دلاله احصائيه عند مقارنتها بالمجموعه المصابه بمرض البول السكري ماعدا كلا من الجلوتاثيون والجلوتاثيون بيروكسيديز زادت زياده ملحوظه

٣- **في كلا من المجموعة المعالجة اولا بالجنسنج قبل الاصابه بمرض البول السكري والمجموعه المعالجه بالجنسنج بعد الاصابه بمرض البول السكري:** قيم كلا من مستوي الجلوكوز الصايم والكوليستيرول والدهون الثلاثية و المالون داي ألدهيد ونشاط ALT و AST و التعبير الجيني لسيتوكروم ب ٤٥٠ ٢ أى ١ زادت زياده ملحوظه بالمقارنه بالمجموعه الضابطه بينما نقصت قيمه كلا من الجلوتاثيون والجلوتاثيون بيروكسيديز وبالعكس عند المقارنه بالمجوعه المصابه بمرض البول السكري نقصت قيم كل المعايير ماعدا الجلوتاثيون والجلوتاثيون بيروكسيديز زادت زياده واضحه .

أوضحت دراسة الهستوباثولوجي :

١ – **في المجموعه الضابطه** : ظهرت خلايا كلا من الكبد والبنكرياس بصوره طبيعيه

٢- **في المجموعه المصابه بمرض البول السكري** :تاثرت خلايا الكبد والبنكرياس تاثيرا شديدا حيث تلفت خلايا الكبد وقلت الانويه فيها وايضا ماتت خلايا البنكرياس وقلت عدد خلايا لانجر هانز .

٣- **في مجموعه الجنسنج** : ظهرت خلايا الكبد والبنكرياس طبيعيه كما في المجموعه الضابطه

٤ – **في كلا من المجموعه المعالجه اولا بالجنسنج قبل الاصابه بمرض البول السكري والمجموعه المعالجه بالجنسنج بعد الاصابه بمرض البول السكري** : حدث تحسن كبيرفي شكل وتركيب خلايا الكبد والبنكرياس مع بعض التغيرات البسيطه بالمقارنه بالمجموعة المصابة بمرض البول السكري.

هناك علاقة ايجابية بين التعبير الجيني لسيتوكروم ب ٤٥٠ ٢ أى ١ وكلا من مستويات الجلوكوز الصايم و الكوليستيرول والدهون الثلاثية و المالون داي ألدهيد ونشاط أنزيمات ALT و AST . في حين أظهر التعبير الجيني لسيتوكروم ب ٤٥٠ ٢ أى ١ علاقه سلبية مع الجلوتاثيون والجلوتاثيون بيروكسيديز.

نستنتج من هذه الدراسة أن :

- تأثير إعطاء مستخلص الجنسنج للفئران المصابة بمرض البول السكري عن طريق الفم ، كان واضحا كمضاد لمرض السكر وحماية خلايا الكبد كذلك تأثيره المضاد لارتفاع الدهون ونشاطه كمضاد للاكسده وعلي هذا يمكن استخدام نبات الجنسنج في معالجة المرضي الذين يعانون من مرض البول السكري نتيجة لتاثيره الجيد المضاد لمرض البول السكري .
- متوسط مستوي الجلوكوز الصايم و وظائف الكبد وقياسات مضادات الاكسده والتعبير الجيني لسيتوكروم ب ٤٥٠ ٢ اي ١ لم تتغير في مجموعة الجنسنج وذلك يدل علي أنه أمن في استخدامه .
- التاثير العلاجي للجنسنج كان اكثر وضوحا من التاثير الوقائي.
- يساعد استخدام تقنية تفاعل البلمره المتسلسل الحديث في تحسين تقدير التعبير الجيني لسيتوكروم ب ٤٥٠ ١٢اي ١.

التوصيات:

١- نوصي بمزيد من الابحاث لربط الجنسنج مع أدوية الكبد الأخري وذلك لتحقيق كفائته الدوائية المفضلة.

٢- أوضح الجنسنج ومكوناته الرئيسية الجنسينوسيد أنشطه مفيده تتضمن دورا هاما في تنظيم وظائف الكبد ومعالجة اضطرابات الكبد من سمية الكبد الحاده والمزمنه والالتهاب الكبدي وتليف وتشمع الكبد وايضا الفشل الكبدي وسرطان الكبد ولذلك نرجو المزيد من البحث عن تاثيرته بالطرق المختلفه المتاحة ليصبح الجنسنج أكثر سلامة وفعاليه .