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THE EFFECTS OF *PANAX GINSENG* ROOT EXTRACT ON CARBOHYDRATE AND LIPID DISTURBANCES ASSOCIATED WITH ALLOXAN-INDUCED DIABETIC RATS

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ABSTRACT

Diabetes is a major public health issue. *Panax ginseng* is an anciently used herbal drug for diabetic treatment. Anti-diabetic effects of *P. ginseng* are attributed to ginsenosides. The objective of current study is to determine the anti-diabetic effects of *Panax ginseng* root extract at gene level. Rats were fed on high-fatty diet for two weeks and divided into three groups (8rats/group): Non-diabetic control group (NDG), Diabetic-group (DG), Diabetic+300mg/kg *Panax ginseng* root extract group (DM+PGE). On 14th day, the rats were kept in overnight-fasting and administered single intra-peritoneal injection of alloxan-monohydrate dissolved in 0.5ml of saline solution at the dose of 120-130mg/Kg body weight (BW). BW and blood glucose were measured on 1st and 14th week. After 14 weeks, fasting/basal blood samples were collected for the biochemical analysis. Liver, skeletal muscle and adipose tissue were also collected for mRNA genes expression of Glucose transporter-4 (GLUT-4), Insulin receptor substrate-1(IRS-1), Insulin receptor (IR), Sterol regulatory element binding protein-1c (SREBP-1c), Fatty acid sythase (FAS), Peroxisome proliferator-activated receptor-α (PPAR-α), Peroxisome proliferator-activated receptor-γ (PPAR-γ) and tumor necrosis factor-α (TNF-α). We found a significant reduction in the BW of DM+PGE group at 14th week. Glycemia was significantly higher in DG and significant reduction was recorded in DM+PGE group. While serum glucose, aspartate aminotransferase (AST), alanine aminotransferase (ALT) and creatinine levels decreased significantly in treated groups. We observed significant increase for catalase (CAT) and decrease for malondialdehyde (MDA) in DM+PGE group in comparison to DG. PGE showed significant increase for HDL-C and decrease for TG. PGE significantly down-regulate hepatic TNF-α. For skeletal muscle, we recorded significant up-regulation for GLUT-4 and PPAR-α while for adipose tissue, we measured up-regulation for IRS-1 and PPAR-γ and significant down-regulation was recorded for TNF-α. In conclusion, PGE has strong anti-diabetic effects mediated by a modulating effect on involved key genes.

Key words: Carbohydrate and lipid metabolism, Diabetes mellitus, *Panax ginseng* root extract, Rats.

INTRODUCTION

Diabetes mellitus is a collection of metabolic defects mainly associated with substantial mortality and morbidity. Diabetes affects approximately 240 million people worldwide and this number is increase substantially to 380 million by 2025. In Pakistan the prevalence of diabetes mellitus is very high, currently, in Pakistan 6.9 million people are affected with diabetes, with projected estimates expected to double by 2025 and affect 11.5 million people (Khuwaja *et al.*, 2004; Khowaja *et al.*, 2007). Diabetes is characterized by insulin resistance, hyperglycemia and oxidative stress also leads to neuropathy, retinopathy, nephropathy, stroke, cardiovascular disease, gum infections, foot amputation polydipsia, polyphagia, polyuria, muscular weakness, weight loss, hyperglycemia and glucosuria (Stitt-Cavanagh *et al.*, 2009). Instead of pharmaceutically agents like insulin, sulfonylurea and biguanides thiazolidinediones which are not health friendly drugs, natural remedies are somehow safer (Lee *et al*., 2010; Cheng *et al.*, 2013). In the past few decades, the interest in the natural hypoglycemic agents, especially those derived from plant sources, has been increased because these herbal products have less toxic side effects than synthetic products on human body (Lee *et al.*, 2010).

Several species of ginseng plant belong to Panax (genus) is one of the widely used herbal medicine in Chinese medicines since ancient time. Therapeutical properties of the ginseng are attributed to triterpene saponins, known as ginsenosides (Kim *et al.*, 2014). Role of ginseng in maintaining body homeostasis, in improving body strength, as remedies for sexual dysfunction, aging, cardiovascular disorders, immune dysfunctioning and cancer have been proven through different biological studies (Attele *et al.*, 2002; Lee *et al.*, 2012). The present project is aimed to explore a complete molecular mechanisms involved in diabetes by evaluating effects root extracts of *Panax*

ginseng (PGE) on several genes and biochemical parameters.

MATERIALS AND METHODS

Experimental Design: Twenty four normoglycemic male adult Wistar rats weighing 150-200 g were selected and acclimatized for 12 days in the animal shed of the University of Veterinary and Animal Sciences (UVAS), Lahore, Pakistan. All the animals were housed in stainless steel cages, 2 animals per cage, with wood litter bedding. Rats were kept in the environmentally controlled room and maintained at 24±5℃ and reverse-dark cycle (12AM-12PM). Rats were given free access to water and standard rat chow (5.8% sunflower oil, 20% dextrose, 22.5% casein 20% dextrose, 22.5% casein, 5.8% sun flower oil, 40.7% maiz starch, 9.7% minerals and 1.3% vitamines by weight) (El-Mesallamy *et al.* 2011). Body weight (BW) was recorded on the first and the last week of the study. Experiment was carried out according to the ethical committee guidelines of the UVAS, Lahore, Pakistan.

After the completion of acclimatization period, rats were given high fat diet (HFD: 31% beef tallow, 28.6% casein, 12.7% maize starch, 3.9% sun flower oil, 6% cellulose, 6.5% dextrose, 9.7% minerals and 1.3% vitamines by weight) for a period of two weeks. Thereafter, rats were randomly divided into three groups (n = 8/group);

- *Non-diabetic group (NDG)*: Rats of this group were served as non-diabetic or negative control and given standard rat chow without supplementation.
- *Diabetic group (DG):* This group included positive control rats (diabetic) and given the standard rat chow without supplementation.
- *DM+PGE group:* This group was comprised of diabetic rats with standard rat chow supplemented with PGE (4% ginsenosides; Hunan Nutramax Inc., China) at the dose of 300 mg/kg/day.

Induction of diabetes: Two weeks after high fat diet feeding, the rats were kept on overnight fasting condition. Thereafter, single intra-peritoneal injection of alloxan monohydrate (Sigma, USA), dissolved in 0.5 ml of saline solution, and was administered at the dose of 120-130 mg/Kg BW. Non-diabetic group received 0.5ml saline solution intra-peritoneally (Ebuehi *et al*., 2010). After 6h of alloxan administration, 20% glucose solution was given to the rats to prevent hypoglycemia (Ebueli *et al.,* 2010). After 3 days of alloxan injection overnight fasting blood samples were collected from tip of tail and glycemia was measured (ACCU check, Germany). Only rats with fasting blood glucose levels higher than 250 mg/dl were considered diabetic and included in the study (Shankar *et al.,* 2005). The duration of experiment was 14 weeks after induction of diabetes.

Blood sampling and biochemical parameters: At the end of experiment, blood sampling was carried out through cardiac puncture and serum was collected after centrifugation. The biochemical analyses of fasting serum glucose (FSG), catalase (CAT), malondialdehyde (MDA), aspartate aminotransferase (AST), alanine aminotransferase (ALT) and creatinine were done using commercially available kits (Randox, UK).

Total serum cholesterol (TC) and triglycerides (TG) were estimated using kits (Bio-Merieux, Marcy-l' Etoile, France). Lipoprotein profile (VLDL-C, LDL-C, HDL-C) were performed through FPLC (AKTA FPLC SYSTEM, GE Healthcare, USA).

Tissue sampling and mRNA expression of genes in the liver, skeletal muscle and visceral adipose tissue: Liver, skeletal muscle and visceral adipose tissues were sampled, cleaned with normal saline water and stored immediately at -80 °C. The RNA was extracted (Laboratoire de Biochimie, MMS, Nantes, France) using trizol reagent (Ambion, USA), according to the manufacturer's instructions. Total RNA concentration was quantified by measuring at 260 nm wavelength. Total RNA (1 µg) was converted into cDNA through the process of reverse-transcribed using Super-Script III Reverse Transcriptase (Invitrogen, France) in a 20µl reaction volume. Quantitaive PCR was performed using SYBR Green Supermix on MyiQ2 Real-Time PCR Detecting system (Bio-Rad, Marnes-la-coquette, France). Primers sequence was determined by the primer3 website. The mRNA levels were normalized using GADPH as a housekeeping gene. Relative quantification was performed using the **ΔΔ**CT method. Primers used for qRT-PCR are listed in Table 1.

Statistical analysis: Results were presented as mean ± S.E.M. Data were analyzed statistically by One way repeated measure analyses of variance (ANOVA) followed by Fisher's PLSD Post hoc test using Statview software (SAS Institute Inc, SAS Campus Drive, Cary, NC, USA), to measure ginseng root extracts effects on all physiological and biochemical parameters except mRNA genes expression in studied organs, which were analyzed by Kruskal Wallis, followed by Fisher's test using Stateview. Significant difference was taken at $P < 0.05$.

Table 1. Genes and primer sequences

Genes	Forward Primer (5'-3')	Reverse Primer (5'-3')
GAPDH	TCCCATTCTTCCACCTTTGATGCT	ACCCTGTTGCTGTAGCCATATTCAT
GLUT4	GCTTCTGTTGCCCTTCTGTC	TGGACGCTCTCTTTCCAACT
IRS-1	GCCAATCTTCATCCAGTTGC	CATCGTGAAGAAGGCATAGG
IR	GTGCTGCTCATGTCCTAAGA	AATGGTCTGTGCTCTTCGTG
SREBP-1c	GGCATGAAACCTGAAGTGGT	TGGGCTTTCACCTGGTTATC
FAS	CGCCGTGGTGCTGGAGATTG	CTTGCCGAGGTTGGTGAGGAAG
PPAR-α	GAGACCCTCGGGGATCTTAG	CGTCTTGTGTCCTGAGCTTG
PPAR-γ	CTGACCCAATGGTTGCTGATTAC	GGACGCAGGCTCTACTTTGATC
TNF-α	GCAGAGCCTTCCAAGCCTACC	GTTACCCAGCCCACCTCCTTTG

Abbreviations: GAPDH (Glyceraldehyde-3-phosphate dehydrogenase), GLUT-4 (Glucose transporter-4), IRS-1 (Insulin receptor substrate-1), IR (Insulin receptor), SREBP-1c (Sterol regulatory element binding protein-1c), FAS (Fatty acid sythase), PPAR-α (Peroxisome proliferator-activated receptor-α), PPAR-γ (Peroxisome proliferator-activated receptor-γ) and TNF-α (tumor necrosis factor-α).

RESULTS

Body weight: A significant ($P < 0.0001$) reduction in the BW of DG was observed compared to NDG on week 14. However, DM+PGE treated group showed significant ($P< 0.0001$) suppression in the reduction of BW (Table 2) compared to diabetic rats.

Blood and fasting serum glucose concentration: We investigated the anti-hyperglycemic activities of PGE in diabetic rats. Random blood glucose level was monitored first at week1 and then at week 14. A significant increase ($P < 0.0001$) in non-fasting blood glucose level was recorded between week1 and week 14 in DG compared with NDG. However, a significant ($P < 0.0001$) reduction in the non-fasting blood glucose concentration was recorded at week 14 in DM+PGE treated group (Table 2) compared to DG.

We also investigated the FSG concentration in the alloxan induced diabetic rats at the end of our experimental trial and recorded significantly ($P < 0.0001$) higher FSG concentration in DG. Treatment of diabetic rats with 300 mg/kg PGE significantly ($P < 0.0001$) reduces the FSG compared to DG rats (Table 3).

Serum lipid Profile: For lipid profile, PGE treated group showed significant ($P < 0.05$) enhancement in HDL-C; significant ($P < 0.0001$) reduction in TG and with no effect on TC, VLDL-C and LDL-C levels. However, DM group significantly increased TC ($P < 0.0001$), VLDL-C ($P < 0.0001$) and LDL-C ($P < 0.0001$) concentrations (Table 3).

Oxidative activity: We found a significant ($P < 0.0001$) decrease in serum CAT and significant increase ($P < 0.001$) for MDA levels in DG. However, DM+PGE group showed significant increase ($P < 0.05$) for CAT and significant decrease ($P < 0.05$) for MDA concentrations (Table 3).

Serum Biochemical parameters: Serum concentrations of creatinine, AST and ALT in NDG, DG and DM+PGE groups are given in Table 3. The PGE showed significant reduction for creatinine ($P < 0.05$), AST ($P < 0.05$) and ALT ($P < 0.05$) concentrations Table 3).

mRNA gene expression in liver: For GLUT-4, IR and PPAR-α no significant changes were observe in diabetic and PGE treated groups. In parallel, we found significant ($P < 0.001$) down-regulation for IRS-1, significant ($P < 0.0001$) up-regulation for SREPB-1c and significant ($P < 0.0001$) down- regulated for FAS in DG and PGE show no effect on these genes. However, we found significant ($P < 0.0001$) down-regulation for TNF-α in DM+PGE group (Fig 1).

mRNA gene expression in skeletal muscles: As shown in Figure 2 a significant down-regulation for IR ($P < 0.0001$), IRS-1 ($P < 0.0001$) and significant up-regulation for TNF-α ($P<0.0001$) in DG were found and treatment show no effect on these genes. However, significant up-regulation for GLUT-4 ($P < 0.001$) and PPAR-α ($P < 0.05$) in DM+PGE treated group was found, whereas, same genes were down-regulated in diabetic rats.

mRNA gene expression in adipose tissues: As shown in figure 3, GLUT-4 and FAS were not affected in any one of the studied groups. However, a significant up-regulation for IRS-1 ($P < 0.001$), PPAR-γ ($P < 0.05$) and significant down-regulation for TNF- α ($P < 0.0001$) in DM+PGE group was found.

Table 2. Effec of *Panax ginseng* on body weight and glucose changes at week 1 and week 14

Parameters	NDG		DG		DM+PGE	
	Week 1	Week 14	Week 1	Week 14	Week 1	Week 14
BW (g)	165.36±1.51	206.20±2.36 γ^{***}	165.93±1.41	143.75±2.20 a^{***}	166.71±0.65	152.12±1.98 b^{***}
Glucose (mg/dl)	83.43±1.28 γ^{***}	83.68±1.35 γ^{***}	417.07±25.43 a^{***}	485.58±25.52 a^{***}	422.16±17.86	227.97±7.04 b^{***}

All values are presented as mean ± S.E.M,
Abbreviations: BW (Body weight), NDG (Non-diabetic control group), DG (Diabetic control group) D+PGE (Diabetic +*Panax ginseng* root extract group)
γ^{***}=P< 0.0001, *Panax ginseng* vs non-diabetic a^{***}=P<0.001, diabetic vs non-diabetic b^{***}=P<0.0001, *Panax ginseng* vs diabetic

Table 3: Effect of *Panax ginseng* on fasting serum glucose, serum lipid concentration (TC, VLDL-C, LDL-C, HDL-C, and TG), Anti-oxidative Stress (CAT and MDA), Creatinine, AST and ALT in alloxan-induced diabetic rats (n=8).

Parameters	NDG	DG	DM+PGE
FSG (mg/dL)	83.68±1.35$^{\gamma***}$	485.58±25.52^{a***}	227.97±7.04^{b***}
TC (g/L)	0.82±0.01 γ^{***}	1.34±0.01^{a***}	1.34±0.02
VLDL-C (g/L)	0.07±0.005 γ^{***}	0.28±0.005^{a***}	0.29±0.005
LDL-C (g/L)	0.06±0.001 γ^{***}	0.55±0.01^{a***}	0.53±0.01
HDL-C (g/L)	0.68±0.01 γ^{***}	0.49±0.008^{a***}	0.54±0.01^{b*}
TG (g/L)	1.25±0.007 γ^{***}	2.12±0.02^{a***}	1.42±0.01^{b***}
CAT (KU/L)	20.83±0.26$^{\gamma*}$	18.60±0.65^{a***}	19.56±0.18^{b*}
MDA (mmol/L)	6.54±0.23	7.55±0.28^{a**}	6.59±0.22^{b*}
Creatinine (mg/dL)	1.63±0.12	2.01±0.03^{a**}	1.8±0.04^{b*}
AST (µ/L)	76.88±1.43 γ^{***}	210.06±3.08^{a***}	195.95±1.13^{b*}
ALT (µ/L)	36.23±0.72	40.9±1.68^{a**}	37.37±0.35^{b*}

All values are presented as mean ± S.E.M
γ^{*}=P< 0.05, *Panax ginseng* vs non-diabetic a^{**}=P<0.001, diabetic vs nondiabetic b^{*}=P<0.05, *Panax ginseng* vs diabetic
γ^{***}=P< 0.0001, *Panax ginseng* vs non-diabetic a^{***}=P<0.0001, diabetic vs nondiabetic b^{***}=P<0.0001, *Panax ginseng* vs diabetic
Abbreviations : NDG (Non-diabetic control group), DG (Diabetic control group) D+PGE (Diabetic +*Panax ginseng* root extract group), FSG (Fasting Serum Glucose), TC (Total Cholesterol), VLDL-C (Very Low Density Lipoprotein-Cholesterol), LDL-C (Low Density Lipoprotein-Cholesterol), HDL-C (High Density Lipoprotein-Cholesterol), TG (Triglyceride), CAT (Catalase), MDA (Malondialdehyde), AST (Aspartate aminotransferase), ALT (Alanine aminotransferase).

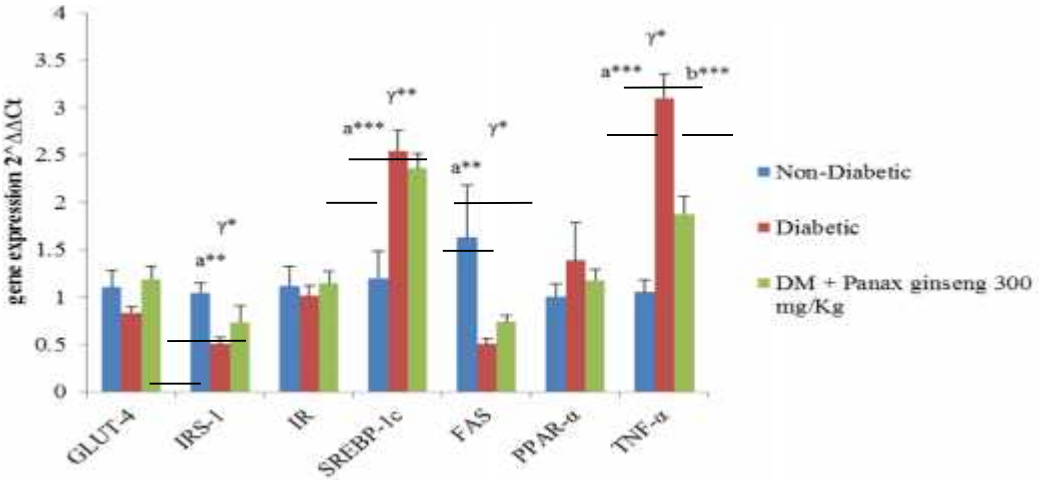

Fig 1. Effect of *Panax ginseng* root extract (PG) on mRNA gene expression of GLUT-4 (Glucose transporter-4), IRS-1 (Insulin receptor substrate-1), IR (Insulin receptor), SPEPB1-c (Sterol regulatory element binding protein-1c), FAS (Fatty acid synthase), PPAR-α (Peroxisome proliferator-activated receptor- α) and TNF-α (tumor necrosis factor-α) in the liver of alloxan induced diabetic male Wistar rats n=8. All values are mean ± S.E.M; γ^{*} P<0.05 and γ^{**} P< 0.001 non-diabetic vs PGE, a^{**} P< 0.001 and a^{***} diabetic P<0.0001 vs non-diabetic, b^{***} P<0.0001 PGE vs diabetic group.

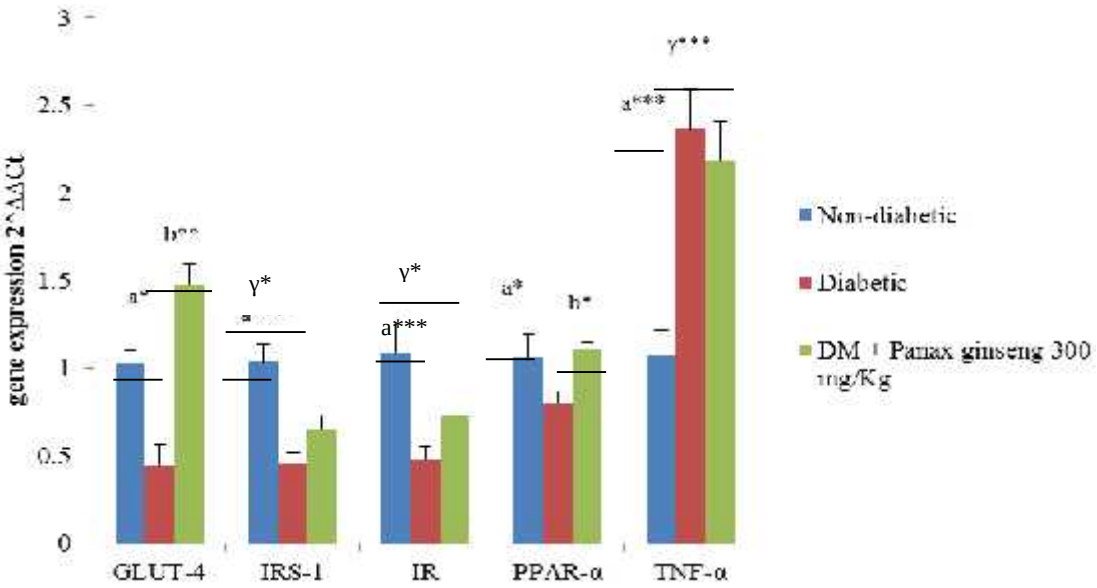

Fig 2. Effect of *Panax ginseng* root extract (PGE) on mRNA gene expression of GLUT-4 (Glucose transporter-4), IRS-1 (Insulin receptor substrate-1), IR (Insulin receptor), PPAR-α (Peroxisome proliferator-activated receptor-α) and TNF-α (tumor necrosis factor-α) in the muscles of alloxan induced diabetic male Wistar rats n=8. All values are mean ± S.E.M. γ* P<0.05 and γ* P<0.0001 non-diabetic vs PGE, a* P<0.05 and a*** P<0.0001 diabetic vs non-diabetic, b* P<0.05 and b** P<0.001 PGE vs diabetic group**

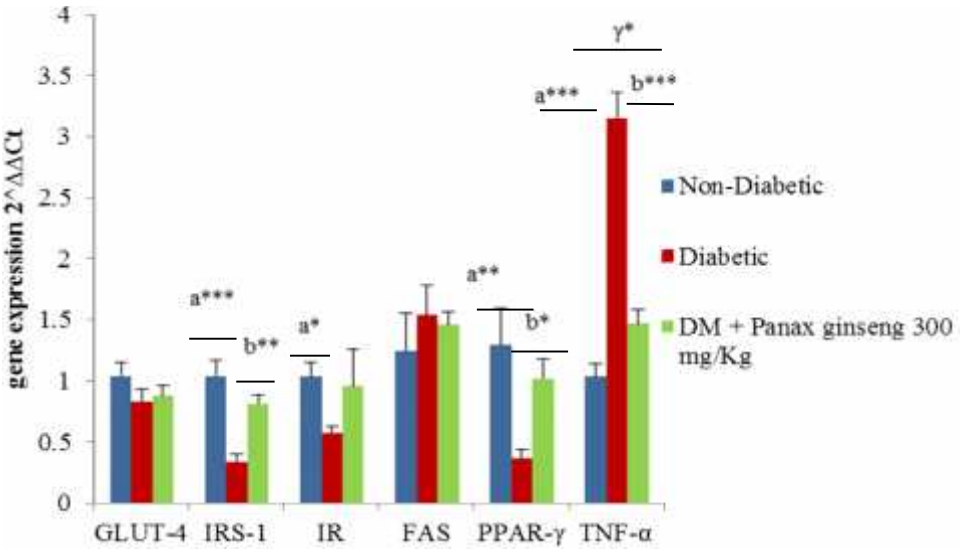

Fig 3: Effect of *Panax ginseng* root extract (PGE) on mRNA gene expression of GLUT-4 (Glucose transporter-4), IRS-1 (Insulin receptor substrate-1), IR (Insulin receptor), Fatty acid synthase (FAS), PPAR-γ (Peroxisome proliferator-activated receptor- γ) and TNF-α (tumor necrosis factor-α) in the adipose tissues of alloxan induced diabetic male Wistar rats. All values are mean ± S.E.M; n=8. γ* P<0.05 non-diabetic vs PGE, a* P<0.05, a P< 0.001 and a*** P<0.0001 diabetic vs non-diabetic, b*P<0.05, b** P<0.001 and b*** P<0.0001 PGE vs diabetic group.**

DISCUSSION

Diabetes is one of the leading health issues. The goal of this study was to check the effects of *P. ginseng*, on diabetic genes and their associated metabolic mechanisms. Medical effects of ginseng are attributed ginsenosides (an active ingredient), present in the leaf, berry and root of ginseng (Attele *et al.*, 2002).

Our results for the BW are in agreement with others (Ebueli *et al.*, 2010; Lee *et al.*, 2012). Reduction in the BW in diabetic state might be attributes to abnormalities in the glucose and lipid metabolism leading

to tissue breakdown and muscle wasting (Baynes *et al.*, 1991).

We also recorded weekly glycemia and found a sustained decrease in blood glucose concentration. Sustained, decreased in hyperglycemia helps to reduce the risk for the development of macro-vascular and micro-vascular complications. The results for FSG are in agreement with other (Shang *et al.*, 2013). To further understand the mechanisms that how PGE act as hypoglycemic agents, we investigated the mRNA expressions of genes involved in the carbohydrate/ glucose and lipid metabolic activities.

In the present study, we found significant up-regulation for GLUT-4 only in muscles after treatment with PGE. It might be possible that diabetic complication caused impairment of insulin action as reflected from reduction in the mRNA expression level of GLUT-4 transporter in liver, muscles and adipose tissues without causing insulin sensitivity in the body. Evidences support the fact that decreased in glucose uptake in insulin resistance was not necessarily correlated with expression of GLUT-4 (Pedersen *et al.*, 1990) and in fact the intrinsic activities of GLUT-4 are involved in this uptake (Konrad *et al.*, 2002).

Interested results for both IR and IRS-1 were found in the present study. In case of IR treatment showed no change for any of the treated group, however, for IRS-1 we reported significant up-regulation only in adipose tissues. IR is a protein to which insulin must bind to carry out its various biological actions in the cells (Calle *et al.*, 2008). Since, the IR is the first effectors of this impairment in insulin signalling. Insulin resistance is associated with down-regulation of both IRS-1 and IRS-2, however the reduction in the IRS-1 level is also associated with age and impaired insulin stimulates glucose uptake (Carvalho *et al.* 2001).

It is well known fact that, dyslipidemia (high TG, LDL-C and low HDL-C) is the main cause of coronary artery disorder and also cause the development of atherosclerosis lesions containing lipid foam cells (Benzi and Morretti, 1995). A significant difference was found only in HDL-C and TG concentrations in treated group. Hypertriglyceridemia is one of the major risk factor for the development of cardio-vascular disorders. The reduction in the TG level is the indication to enhance energy expenditure of the whole body. Thus it is important to reduce hypertriglyceridemia in diabetic state. We studied the genes involved in the lipid metabolism to better understand that how PGE plays a role to correct the dyslipidemia and hypertriglyceridemia in diabetic state.

Liver plays important role in metabolic activities of the body, since it serves as a main storage releasing site for carbohydrate and fatty acid synthesis (Quan *et al.*, 2012), SREBP-1c plays a pivotal role lipogenic transcriptional factor and lipolytic genes (Yuan *et al.*, 2011). Our results for SREBP-1c in treated group are not in agreement with other (Quan *et al.*, 2012).

It is believed that FAS catalyzes the biosynthesis of FA particularly in the liver and adipose tissue. Since, the gene expression of FAS is predominantly regulated by nutritional and hormonal signals (Menendez *et al.*, 2009). The results for FAS are in agreement with other researchers (Banz *et al.*, 2009).

Our data proved that ginseng have an effective role to improve lipid metabolism through up regulation of PPAR-α in muscles and PPAR-γ in adipose tissues. PPAR-α leads to improves the lipid profile and also insulin sensitivity by enhancing the β-oxidation of fatty acids (Banz *et al.*, 2009), thus dysfunctioning of PPAR-α results the metabolic abnormalities in TC and TG by causing impairment in fatty acid and lipoprotein metabolism (Yoon *et al.*, 2003). PPAR-γ are widely distributed in the adipocytes; regulates the expression of key genes involved in the glucose and lipids metabolism and also plays an important role in the induction of adipocytes differentiation (Lee *et al.*, 2010) and stimulates the production of small insulin signaling pathway by inducing the expression of genes involved with the insulin signaling cascade (Banz *et al.*, 2009).

Hyperglycemia is one of the major contributing factors to increase ROS production in diabetes; thence the body suffers with oxidative stress and causes severe tissue damage and consequently pathogenesis of vascular diseases which are the main cause of morbidity and mortality in diabetic state. In the present study, we found that the body of diabetic rats suffers with oxidative stress. Evidences support the fact that alloxan generates the free radicals and destroyed the pancreatic β-cells. Although anti-oxidant enzymes CAT and MDA have an important role to neutralize the toxicity of active oxygen, however, reduction in anti-oxidant enzymes has been seen in diabetes (West, 2000). Anti-oxidant activity (scavenging activities) of ginseng is due to their active ingredient ginsenosides, having powerful role scavenging activities by stimulating gene expression of anti-oxidant enzymes and thus enhancing their activities (Surh *et al.*, 2001). MDA is an important biomarker of oxidative stress in the body and it is the final product of lipid break down (Ren *et al.*, 2013). CAT an enzyme that plays a vital role to protect the cells against toxic effects of hydrogen peroxide. The main catalytic activity of CAT enzyme is to decompose hydrogen peroxide to water and oxygen (Kodydkova *et al.*, 2014).

TNF-α has been proposed a link between adiposity and the development of insulin resistance. Further studies revealed that the concentration of plasma TNF-α could be linked with abdominal adiposity. TNF-α is an important contributor to systemic insulin resistance by impeding insulin's actions in liver and skeletal muscle. Oxidative stress and inflammation plays a major role in the development of chronic diseases (Camps and

Garcia-Heredia, 2014); causes adipocyte lipolysis, release of free fatty acids, exacerbating insulin resistance, metabolic risk factors and finally diabetes (Badish *et al.*, 2010). The results for TNF-α is in agreement with other (Gu *et al*., 2013).

Creatinine is a waist product of the body which must be excreted via kidneys through glomerular filtration. Increased in blood creatinine level indicates impaired renal function (Salih, 2012). The stability of creatinine level in after treatment indicates the protective effects against alloxan toxicity of this natural remedy. There is strong evidence to support the fact that liver enzymes AST and ALT are associated with increase the risk of diabetes and considered as an important liver marker to identify liver diseases (Kunutsor *et al*., 2013). We reported a significant reduction for both AST and ALT in treated group.

In conclusion the root extract of *P. ginseng* have strong anti-hyperglycemic, anti- hypertriglyceridemic and anti-oxidative properties. It also shows strong influence on the activation on the expression of genes involved in the metabolic pathways of lipid and glucose which previously become dysfunctional in diabetic state.

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