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Research Article

A NOVEL POLYHERBAL PREPARATION FOR THE MANAGEMENT OF TYPE-2 DIABETES MELLITUS: A CLINICAL STUDY

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ABSTRACT

Several studies have used medicinal plants for the management of experimental and clinical diabetes but a novel polyherbal preparation which contains plants having anti-diabetic and aphrodisiac properties were used in the present study for the management of type-2 diabetic patients. Mild Diabetic group (MD) (n=52) fasting blood glucose ranges from 100-200mg/dl and postprandial up to 300mg/dl and Severe Diabetic group (SD) (n=41) fasting blood glucose level above 200mg/dl and postprandial level above 300mg/dl of both sex who gave written consent to participate in the study were enrolled and received polyherbal preparation (PHP) treatment for 12 weeks at a dose of 1g three times a day after food for MD and 1.5g three times a day after food for SD subjects. Fasting and postprandial blood glucose and HbA_{1c} levels of MD and SD were significantly decreased after 12 weeks of PHP treatment ($p<0.001$), while, total hemoglobin was significantly increased ($p<0.001$). Serum insulin and C-peptide concentrations of both MD ($p<0.005$) and SD ($p<0.012$) groups were decreased significantly after 12 weeks of PHP treatment whereas, no change was recorded in estradiol levels in MD and SD patients. However, significant increase in testosterone levels of both MD ($p<0.005$) and SD ($p<0.012$) men was recorded. Resistin, MCP-1 and TNF α concentrations of MD ($p<0.005$) and SD ($p<0.012$) was significantly decreased at 12 weeks of treatment. In conclusion, polyherbal preparation used in the present study has potential anti-diabetic property by reducing blood glucose level and favouring restoration of cytokine network and serum hormones towards physiological adverse range without any obvious adverse effects.

Keywords: Diabetes mellitus, Polyherbal preparation, Medicinal plants, Cytokines, Sex steroids.

INTRODUCTION

Diabetes mellitus is a metabolic disease characterized by hyperglycemia resulting from defects in insulin secretion or action or both.[1] It is a major cause of disability and hospitalisation in recent years. Prevalence of diabetes is increasing in all countries, especially in India, at an alarming rate.[2] Though a sub-section of type-2 diabetic patients can be managed by diet alone, most patients require oral hyoglycemic agents and/or insulin. Insulin therapy affords effective glycemic control, yet its shortcomings such as ineffectiveness on oral administration, short half-life, requirement of constant refrigeration and in the event of excess dosage fatal hypoglycemia limit its usage.[3] Treatment with sulfonylureas and biguanides are also associated with adverse effects.[4] Contrary to this, many traditional treatments have been recommended in the alternative system of medicine for treatment of diabetes mellitus. Several medicinal plants have been proposed for the treatment of diabetes mellitus and claimed to have anti-hyperglycaemic effects. Studies have demonstrated the anti-hyperglycemic activity of *Terminalia Arjuna, Eugenia jambolana*, *Trigonella foenum graecum*, *Gymnema sylvestre*, *Withania somnifera*, *Momordica charantia* and *Mucuna pruriens*.[5-8]

It has been proved beyond doubt that androgens have an important role in maintaining glucose homeostasis and diabetes mellitus has adverse effects on the androgenic status in men as well as animal models.[9] In the current situation, male diabetic subjects often become impotent due to deficiency in circulating testosterone.[9,10] In accordance with this, our previous experimental study showed that, insulin replacement partially prevents diabetes-induced atrophic changes in testis. The partial restoration of body weight observed in insulin-replaced group of rats was found to be due to maintenance of growth hormone, thyroid hormones and testosterone levels partially or completely.[11-13]

It has been reported that in animal models, sex hormones are important regulators of insulin-mediated events in skeletal muscles.[10,14] In particular, insulin- stimulated glucose uptake and glycogen synthesis were influenced by sex steroids. Therefore, plants having aphrodisiac property have been included along with anti-diabetic plants for the preparation of a novel polyherbal antidiabetic agent. In addition, the authors published an original research article Ismail Khan et al. reported that, efficacy of the polyherbal preparation (PHP) tested in experimental diabetic male albino rats showed significant reduction in blood glucose levels and free of any adverse effects.[15] Based on this background, again PHP is used in the present clinical study for the management of type-2 diabetes mellitus.

MATERIALS AND METHODS

Polyherbal preparations

The specific parts of six dried medicinal plant parts *i.e, Gymnema sylvestre* leaves, *Eugenia jambolana* seeds, *Momordica charantia* fruits, *Trigonella foenum graecum* seeds, *Withania somnifera* roots and *Mucuna pruriens* seeds were purchased from local market and were authenticated by Dr S. Shahul Hameed (Principal, Government Unani Medical College, Chennai, India) and specimens were preserved in the department of (Ilmul Adwia) Pharmacology. Above mentioned plant parts were taken in equal ratio and they were ground in a mixer to make fine powder and filled in 500/750mg capsules. The bioactive compounds and active principles which are present in the medicinal plants are not lost or substandard while preparing polyherbal preparation as per the procedure of Indian system of medicines.

A single centered, open labeled non-randomized clinical trial conducted during the period of November 2008-June 2011 on people with diabetes of both gender approached for alternative/herbal treatment aged between 35-65 years were recruited from outpatient clinic of Thousand lights Diabetes centre.

Patients with symptoms of diabetes and who had not received any anti-diabetic drugs was screened and divided into two groups based on their blood glucose levels. Mild Diabetic group (MD) (n=52): fasting blood glucose ranges from 100-200mg/dl and postprandial (pp) 180-300mg/dl. Severe Diabetic group (SD) (n=41): fasting blood glucose level above 200mg/dl and postprandial level above 300mg/dl. Patients who gave written consent to participate in the study were enrolled and received polyherbal preparation (PHP) treatment for 12 weeks at a dose of 1g three times a day after food for MD and 1.5g three times a day after food for SD subjects. Patients with other systemic illness, recently hospitalized, pregnancy and lactating women were not included in the study.

The study was conducted in compliance with "Ethical Guidelines for Biomedical Research on Human subjects ICMR 2000" for conduct of

trial with herbal remedies. The study protocol was approved by Institutional Human Ethical Committee (IHEC) PGIBMS/CO/Human Ethical/2008-09/206 dt 06 Aug 2008).

At monthly visits, patients were followed up with detailed clinical examination, assessment of compliance to drug intake (including counting of unused capsules), diet adherence, record of any adverse events, fasting and PP blood glucose levels and safety investigations. In addition to this, following parameters were performed before and after 12 weeks of PHP treatment. Fasting and postprandial (PP) blood glucose levels were estimated by hand held glucometer from Abott, USA. For safety measures total RBC, WBC, PCV, MCH and Hb were assessed with an auto-analyzer (Erba Chem 5 plus) Trans Asia, USA. Liver function test: SGOT and SGPT were estimated by IFCC/kinetic method [17,18] and ALP (Alkaline phosphatase) was detrermined by GSCC/kinetic method.[19] Blood urea was estimated by GLDH/kinetic method.[20,21] Serum creatinine was determined by alkaline picrate method.[22,23] Hb A_{1C} Glycosylated haemoglobin) was determined using Biosystems micro columns.[16] Serum insulin, C-peptide, testosterone and estradiol were estimated using RIA kits from Diasorin, Italy. Insulin sensitivity index (HOMA) is a calculated value using fasting serum insulin and blood glucose levels. Cytokines like resistin was estimated by ELISA kit from Orgenium, Finland and adiponectin, MCP-1 and TNF α were estimated by ELISA kits from Raybiotech, USA. The data were subjected to statistical analyses.

Data analysis

Values are expressed as mean ± standard deviation. Statistical significance was calculated by using student's t test. Paired't' test was used to compare the baseline and end point biochemical values and Wilcoxon signed Ranks Test was used for serum hormones and cytokines values. In the present study, statistical significance was set at $p<0.05$

RESULTS

In MD group (n=52) at the end of 4weeks, 2 men and in 8 weeks, 2 men and 2 women were withdrawn from the study because blood glucose levels were decreasing below 80mg/dl. Following Tables (1, 1a and 1b) were baseline characteristics of bio-chemical, serum hormones and cytokines levels of Mild and Severe diabetic subjects.

Table 1: Baseline characteristics and bio-chemical values of Mild and Severe diabetic patients

	Mild Diabetes(MD)	Severe Diabetes(SD)	p value
n (male/female)	**52(27/23)**	**41(22/19)**	
Age (years)	49 ± 6	50 ± 8	0.779
BMI(Kg/m2)	25 ± 5	25 ± 4	0.41
Fasting glucose(mg/dl)	146 ± 28	267 ± 36	0.0001
PPBG(mg/dl)	277 ± 36	404 ± 41	0.0001
HbA1c	7.7 ± 0.4	9.5 ± 0.3	0.0001
RBC	4.6 ± 0.5	4.7 ± 0.6	0.77
WBC	7.2 ± 1.8	7.3 ± 1.4	0.67
Haemoglobin	11.7 ± 1.1	9.4 ± 0.8	0.0001
PCV	40 ± 3	39 ± 5	0.23
MCV	89 ± 5	82 ± 11	0.92
MCH	28 ± 3	27 ± 2	0.13
SGOT	21 ± 6	20 ± 7	0.12
SGPT	15 ± 3	16 ± 7	0.72
ALP	83 ± 12	91 ± 18	0.65
Urea	15 ± 2.4	14 ± 4	0.14
Creatinine	0.84 ± 0.15	0.91 ± 0.18	0.51

Each value represents Mean ± SD. Unpaired't' test was used to assess the values between Mild and Severe diabetic patients.

Table 1a: Baseline values of serum hormones and cytokines levels of Mild and Severe diabetes patients.

	Mild Diabetes	Severe Diabetes	p value
n (male/female)	30(15/15)	30(15/15)	
Insulin (uIU/ml)	15 ± 3	21 ± 4	0.0001
C-peptide	2.62 ± 0.43	3.4 ± 0.82	0.0001
HOMA	4.7 ± 0.64	9.7 ± 0.9	0.0001
Adiponectin	3.8 ± 0.67	2.1 ± 0.48	0.0001
Resistin	21 ± 4	24 ± 6	0.216
MCP-1	581 ± 62	642 ± 76	0.068
TNFα	1.66 ± 0.21	1.8 ± 0.26	0.274

Each value represents Mean ± SD. Unpaired't' test was used to assess the values between Mild and Severe diabetic patients.

Table 1b: Baseline values of serum hormones and cytokines levels of Mild and Severe diabetes patients.

	Mild Diabetes (MD)	Severe Diabetes (SD)	p value
n (male/female)	30 (15/15)	30 (15/15)	(male/female)
Testosterone	3.35±1.3 / 0.43±0.3	2.2±0.7 / 0.40±0.5	0.05 / 0.78
Estradiol	41±16 / 133±78	39±17 / 110±66	0.82 / 0.13

Each value represents Mean ± SD. Unpaired't' test was used to assess the values between Mild and Severe diabetic patients.

Blood glucose, haemoglobin level and safety measures

After 12 weeks of PHP treatment, the mean fasting and postprandial blood glucose levels and HbA_{1c} in MD and SD group was decreased significantly whereas significant increase in total haemoglobin level was observed. Whereas, Toxicological parameters like (Haemogram) RBC, WBC, PCV, MCV, MCH, Liver Function Test (LFT) SGOT,SGPT, ALP and Kidney Function Test (KFT) Urea and creatinine were unaltered within the groups from baseline to end of the study period in MD and SD groups (Table 2 and 3).

Table 2: Effect of PHP on bio-chemical parameters of Mild diabetic patient

	Mild Diabetes	Mild Diabetes after 12 weeks	p value
n (male/female)	52 (27/23)	46 (25/21)	
Fasting glucose(mg/dl)	146 ± 28	95 ± 17	0.0001
PPBG(mg/dl)	277 ± 36	152 ± 31	0.0001
HbA1c	7.7 ± 0.4	5.7 ± 0.6	0.0001
RBC	4.6 ± 0.5	4.5 ± 0.4	0.87
WBC	7.2 ± 1.8	6.3 ± 2.3	0.63
Haemoglobin	11.7 ± 1.1	13.7 ± 0.8	0.0001
PCV	40 ± 3	39 ± 7	0.76
MCV	89 ± 5	85 ± 7	0.78
MCH	28 ± 3	30 ± 2	0.44
SGOT	21 ± 6	22 ± 5	0.06
SGPT	15 ± 3	14 ± 5	0.35
ALP	83 ± 12	88 ± 21	0.70
Urea	15 ± 2.4	14 ± 3.2	0.20
Creatinine	0.84 ± 0.15	0.92 ± 0.16	0.18

Each value represents Mean ± SD. Paired't' test was used to assess the values between baseline and end of the study

Table 3: Effect of PHP on bio-chemical parameters of Severe diabetic patient.

	Severe Diabetes	Severe Diabetes after 12 weeks	p value
n (male/female)	41 (22/19)	36 (20/16)	
Fasting glucose(mg/dl)	267 ± 36	164 ± 29	0.0001
PPBG(mg/dl)	404 ± 41	231 ± 39	0.0001
HbA1c	9.5 ± 0.3	7.5 ± 0.4	0.0001
RBC	4.7 ± 0.6	4.8 ± 0.6	0.52
WBC	7.3 ± 1.4	6.9 ± 2.5	0.71
Haemoglobin	9.4 ± 0.8	11.9 ± 0.7	0.0001
PCV	39 ± 5	40 ± 7	0.84
MCV	82 ± 11	76 ±13	0.74
MCH	27 ± 2	31 ± 3	0.23
SGOT	20 ± 7	19 ± 7	0.97
SGPT	16 ± 7	14 ± 4	0.57
ALP	91 ± 18	94 ± 19	0.71
Urea	14 ± 4	14 ± 5	0.43
Creatinine	0.91 ± 0.18	0.98 ± 0.11	0.50

Each value represents Mean ± SD. Paired't' test was used to assess the values between baseline and end of the study

Serum hormones, adipokines and inflammatory cytokines estimation

Serum insulin and c-peptide concentrations of both MD and SD group were decreased significantly after 12 weeks of PHP treatment. And in estradiol levels of both MD and SD group of both men and women did not differ from baseline to end of the study period between the groups. Whereas serum testosterone level was significantly increased in men of MD and SD group from baseline to the end of the study respectively but no significant changes was recorded in women of MD and SD group. Resistin, MCP-1 and TNF-α concentration of MD and SD group at baseline of the study was significantly decreased at 12 of PHP treatment. But adiponectin level was significantly increased from baseline of the study to 12 weeks in MD & SD subjects are summarized in (Table 4a, 4b, 5a and 5b).

Table 4a: Effect of PHP on Serum hormones and cytokines of Mild diabetic patient

	Mild Diabetes	Mild Diabetes after 12 weeks	p value
Insulin (uIU/ml)	15 ± 3	7.7 ± 0.86	0.005
C-peptide	2.62 ± 0.43	1.8 ± 0.21	0.005
HOMA	4.7 ± 0.64	2.26 ± 0.51	0.005
Adiponectin	3.8 ± 0.67	7.6 ± 1.2	0.005
Resistin	21.37 ± 4.1	14.5 ± 5.2	0.005
MCP-1	581 ± 62	398 ± 73	0.005
TNFα	1.66 ± 0.21	1.05 ± 0.25	0.005

Each value represents Mean ± SD. Wilcoxon signed Ranks Test was used to assess the values between baseline and end of the study

Table 4b: Effect of PHP on Serum hormones and cytokines of Mild diabetic patient

Sex	Mild Diabetes (male / female)	Mild Diabetes after 12 weeks (male / female)	p value (male / female)
Testosterone	3.35 ± 1.3 / 0.43 ± 0.3	5.2 ± 0.7 / 0.34 ± 0.28	0.005 / 0.67
Estradiol	41 ± 6 / 153 ± 78	39 ± 16 / 158 ± 82	0.82 / 0.61

Each value represents Mean ± SD. Wilcoxon signed Ranks Test was used to assess the values between baseline and end of the study

Table 5a: Effect of PHP on Serum hormones and cytokines of Severe diabetic patient.

	Severe Diabetes	Severe Diabetes after 12 weeks	p value
Insulin (uIU/ml)	21 ± 4.3	17 ± 2.4	0.012
C-peptide	3.4 ± 0.82	2.1 ± 0.4	0.012
HOMA	9.7 ± 0.9	4.6 ± 0.53	0.012
Adiponectin	2.1 ± 0.48	4.5 ± 0.51	0.012
Resistin	24 ± 5.6	17.3 ± 3.9	0.012
MCP-1	642 ± 76	490 ± 77	0.012
TNFα	1.8 ± 0.26	1.2 ± 0.17	0.012

Each value represents Mean ± SD. Wilcoxon signed Ranks Test was used to assess the values between baseline and end of the study

Table 5b: Effect of PHP on Serum hormones and cytokines of Severe diabetic patient.

	Severe Diabetes	Severe Diabetes after 12 weeks	p value
n (male/female)	30 (15/15)	30 (15/15)	(male/female)
Testosterone	2.2 ± 0.7 / 0.40 ± 0.5	3.8 ± 0.8 / 0.27 ± 0.22	0.012 / 0.461
Estradiol	39 ± 17 / 110 ± 66	39 ± 16 / 114 ± 78	0.57 / 0.77

Each value represents Mean ± SD. Wilcoxon signed Ranks Test was used to assess the values between baseline and end of the study

DISCUSSION

Diabetes mellitus is a metabolic disorder and its consequences affect various systems of human physiology, and hence, it requires multifunctional or synergistic effects of medicinal plants for the management. The plant parts used in the polyherbal preparation have been used since ancient times by the physicians of traditional medicine including Unani system of medicine as anti-diabetic drugs. Since it is a polyherbal preparation it has a multiple and synergistic effects on various systems of human body.

The mean fasting and postprandial blood glucose levels of MD subjects after 12 weeks of polyherbal treatment were brought back to almost normal range, however in SD subjects it was restored partially. Sahana et al. [24] reported that the treatment with C*occinia cordifolia* extract in newly detected type-2 diabetic patients for 90 days results in 16% decrease in fasting blood glucose level and 18% in PP blood glucose level. Several studies of medicinal plants claimed to have significant reduction in blood glucose level, [5] but in the present study PHP restores the blood glucose level to normal range. Hence, the efficiency of PHP is better than single medicinal plant treatments. In the present study, HbA_{1C} percentage was significantly decreased in both mild and severe diabetic groups after 12 weeks of polyherbal preparation (PHP) treatment suggesting that there is a reduction of generalized glycosylation of proteins in circulation. Glucosuric symptoms were also controlled in both mild and severe diabetic groups. However, there were episodes of hypoglycemia recorded often in MD group.

While screening for any toxicological effects of PHP, no change in haematological parameters was recorded from baseline to 12 weeks of the study period except haemoglobin level which was significantly increased in both MD & SD groups of patients. Further, liver function test (LFT) and kidney function test (KFT) values observed had no deviation from baseline to 12 weeks of the study period. Thus, oral administration of PHP had no adverse effects in diabetic patients and in our previous study, 500mg of PHP /kg b.w. does not results in any adverse effects in the experimental diabetic rats [15] and the same also claimed by the pharmacology of Indian system of medicines.

In the present study serum insulin levels were significantly decreased in both mild and severe diabetic groups after 12 weeks of PHP treatment, thereby insulin sensitivity index also well maintained in both groups. In accordance with this, previous reports regarding medicinal plants and its constituents are very well known in regulating β-cell function and insulin levels in experimental diabetic animals.[15,25] Previous studies have shown that elevated serum levels of C-reactive protein and several cytokines are associated with obesity, insulin resistance and type-2 DM. In the adipose tissue, there is a local production of TNF-α and IL-6 both of which have potent insulin antagonistic properties. [26] Zhuang et al. [27] reported that circulating levels of adiponectin display a strong association with insulin resistance and body fat, and experimental data suggest that adiponectin can reduce hyperglycemia, insulin resistance, inflammation, atherosclerosis and potentially adiposity.

Inflammatory cytokines like TNF α, MCP-1 and IL-6 are increased in diabetes mellitus when compared with healthy subjects.[26] In the present study, 12 weeks of PHP treated MD & SD subjects showed a significant decrease in TNF α and MCP-1 levels suggesting that PHP acts as an anti-inflammatory drug thereby reducing inflammatory cytokines in the circulation. Circulating resistin level was significantly increased in diabetic subjects but the adiponectin level was reduced when compared with healthy subjects. [27] In the present study, adiponectin and resistin were well regulated after 12 weeks of PHP treatment in both MD & SD subjects.

It is well established that sex steroids play a vital role in glucose homeostasis. Impotency is one of the complications in diabetic men which sets in a large section of diabetic subjects. A systemic review by Ding et al. [28] of 43 studies including 6427 men suggested that higher testosterone plasma levels was associated with lower risk of type-2 DM and *vice versa*. Fukai et al. [29] demonstrated that serum testosterone levels are lower in a large number of Japanese patients with type-2 DM when compared with healthy men. Grossman et al. [30] found that 43% of men with type-2 DM have reduced total testosterone, whereas 57% had reduced calculating free testosterone. Testosterone production was severely hampered by diabetes mellitus in men. In the present study, after 12 weeks of PHP treatment there was a significant increase in serum testosterone concentration in both mild and severe diabetic men. However, no significant change was recorded in circulating estradiol concenteration of both the genders of mild and severe diabetic subjects. Since PHP contains aphrodisiac properties it may not have an impact on sex steroidogenesis of women, hence no significant change was recorded in serum estadiol concentrations. Both *Withania somnifera* roots and *Mucuna prurien* seeds are potent aphrodisiac plant parts as per the pharmacology of Indian system of medicines.

It is proposed that the active principles, bioactive compounds and fibers present in the medicinal plants of PHP may play a vital role in regulating the blood glucose level by acting both at pancreatic and extra pancreatic levels. Similarly, Flavanoids isolated from *Eugenia Jambolana* seeds stimulated 16% increase in insulin release *in vitro* from pancreatic cells.[31] 10 and 25 nM Momordin extract from *Momordica charantia* significantly increased the expression of PPAR delta mRNA 1.5 fold and PPAR delta promoter activity in a dose-dependant manner reaching more than 1.5 fold relative to control.[32] Intestinal disaccharidase activity and glucose absorption were decreased and gastrointestinal motility was increased by the soluble dietary fiber fraction of *Trigonella foenum graecum* and thereby suppressed the elevation of blood glucose after oral glucose ingestion in both non-diabetic and type-2 diabetic rats. Activation of

AMPK in muscle is known to increase the expression of GLUT4 and increased plasma membrane GLUT4 has been measured in the skeletal muscle of insulin-resistant mice treated with *Momordica charantia* extracts.[33] *Momordica charantia* extracts administered in insulin-deficient rats were shown to boost hepatic activity of glucose-6-phosphate dehydrogenase, while reducing the concentration of glucose-6-phosphate.[34] Metformin has been reported to have similar effect on hepatic enzyme activity.[35] Treatment with 250 and 500 mg/kg b.w. of aqueous ethanolic extract of *Tragia involucrata* to type 2 diabetic rats showed significant antidiabetic and hypolipidemic potential in dose dependant manner which could be used as the supportive treatment of diabetes mellitus for better glycemic control.[36] Adminsitration of aqueous extract of *Momordia charantia* fruit in diabetic mice results in decreased insulin resistance as a result of increased GLUT4 protein content in plasma membrane of muscle.[37] Ethanolic extract (200mg/kg b.w.) of *Gymnema sylvestre* leaves exert potent hypoglycemic effect in experimental diabetic rats.[5] Oral administration of aqueous extract of *Mucuna pruriens* seeds at a dose of 200mg/kg b.w. significantly reduced the glucose level to normal range in experimental diabetic rats.[8] Ethanolic extract of *Withania somnifera* root prevents glucose mediated collagen glycation and cross linking *in vitro* suggesting its therapeutic role in the prevention of glycation-induced pathogenesis in diabetes mellitus and aging.[38] Since the above discussed medicinal plants are constituents of PHP in the present study the antidiabetic properties recorded in type-2 diabetic subjects may be the synergistic and cumulative effect of the medicinal plants. It is also obvious that the effect of PHP is relatively better than the individual effect of these plants. However, the limitation of the present study was not a placebo controlled or randomized trial.

CONCLUSION

It is concluded from the present study that polyherbal preparation used in the present study has potential anti-diabetic property by reducing blood glucose level and favouring restoration of cytokine network and serum hormones towards physiological range without any obvious adverse effects.

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REFERENCES

1. James R. Members of the expert committee on the diagnosis and classification of Diabetes Mellitus. Dia Care 2005; 28: 37-42.
2. Rebecca K, Ramaswamy R, Ganapathi B, Anura VK. Effect of supplementation of *Coccinia cordifolia* extract on newly detected diabetic patients. Dia Care 2008; 31: 216–20.
3. Grover JK, Vats V, Rathi SS. Anti-hyperglycemic effect of *Eugenia jambolana* and *Tinospora cordifolia* in experimental diabetes and theireffects on key metabolic enzymes involved in carbohydrate metabolism. J Ethnopharmacol 2000; 73: 461-70.
4. Khan CR, Shecter Y. Insulin, oral hypoglycemic agents and the pharmacology of the endocrine pancreas. In: Gilman AG, Rall TW, Nies AS, Taylor P, eds, Goodman and Gilman's The pharmacological basis of therapeutics. 8th ed. Vol. 2. New York: Pergamon Press, Maxwell Macmillan International editions, 1991: 1463-95.
5. Yadav M, Lavania A, Tomar R, Prasad GB, et al. Complementary and comparative study on hypoglycemic and antihyperglycemic activity of various extracts of *Eugenia jambolana* seed, *Momordica charantia* fruits, *Gymnema sylvestre*, and *Trigonella foenum graecum* seeds in rats. Appl Biochem Biotechnol 2010; 160: 2388-400.
6. Kaushik G, Satya S, Rakesh Kumar K, Naik SN. Commonly consumed Indian plant food materials in the management of diabetes mellitus. Dia & Metab Synd: Clin Res & Rev 2010; 4: 21-40.
7. Udayakumar R, Kasthurirengan S, Mariashibu TS, Rajesh M, et al. Hypoglycaemic and Hypolipidaemic effects of *Withania somnifera* root and leaf extracts on alloxan-induced diabetic rats. Int J Mol Sci 2009; 20: 2367-82.
8. Alam Morshed M, Anwarul Haque, Begum Rokeya, Liaquat A. Anti-hyperglycemic and lipid lowering effect of terminalia arjuna bark extract on streptozotocin indiced type 2 diabetic model rats. Int J Pharm Pharm Sci 2011; 3: 450-454.
9. Abdulmaged MT, Farid Saad, Andre Guay. The dark side of testosterone deficiency: II. Type 2 Diabetes and Insulin Resistance. J Androl 2009; 30: 23-32.
10. Livingstone C, Collison M. Sex steroids and insulin resistance. Clin Sci 2002; 102: 151-6.
11. Boujon CE, Bestetti GE, Abramo F, et al. The reduction of circulating growth hormone and prolactin in streptozotocin-induced diabetic male rats is possibly caused by hypothalamic rather than pituitary changes. J Endocrinol 1995; 145: 19-26.
12. Sudha S, Ravisankar B, Valli G, et al. Streptozotocin-diabetes impairs prolactin binding to Leydig cells in prepubertal and pubertal rats. Horm Metab Res 1999; 31: 538-86.
13. Sudha S, Ravisankar B, Julie P.M, et al. Influence of streptozotocin-induced diabetes and insulin replacement on the pituitary-testicular axis during sexual maturation in rats. Exp Clin Endocrinol Diab 2000; 108: 14-20.
14. Duccluzeau PH, Fletcher LM, Vidal H, et al. Molecular mechanisms of insulin-stimulated glucose uptake in adipocytes. Dia Metab 2002; 28: 85-92.
15. Ismail khan A, Yuvaraj S, Suthagar E, et al. Studies on hypoglycaemic effects of polyherbal preparation in streptozotocin- induced diabetic male albino rats. Hum & Exp Toxicol 2009; 28: 679-87.
16. Bisse E, Abraham EC. New less temperature-sensitive microchromatographic method for the separation and quantitation of glycosylated hemoglobins using a non-cyanide buffer system. J Chromatogr 1983; 344: 81-91.
17. Thefeld W, Hoffmeister H, Busch EW, et al. Referenzwertte fur die Bestimmungender Transaminasen GOT and GPT sowie der alkalischen Phosphatase im Serum mit optimiertten Standardmethoden. Dtsh Med Wochenschr 1974; 99: 343-51.
18. Bergmeyer HU, Scheibe P, Wahlefeld AW. Optimization of methods for aspartate aminotransferase and alanine aminotransferase. Clin Chem 1978; 24: 58-73.
19. Bretaudiere JP, Vassault A, Amsellem L, Pourci ML, et al. Criteria for establishing a standardized method for determining alkaline phosphatase activity in human serum. Clin Chem Acta 1977; 23: 2263-74.
20. Hallett CJ, Cook JG. Reduced nicotinamide adenine dinucleotide-coupled reaction for emergency blood urea estimation. Clin Chem Acta 1971; 35: 33-37.
21. Kaltwasser H, Schlegel HG. NADH-dependent coupled enzyme assay for urease and other ammonia-producing systems. Ann Biochem 1996; 16: 132-38.
22. Tietz NW. Fundamentals of Clinical Chemistry. 2nd ed WB Saunders Co. Philadelphia P.A. (1976) 47-51.
23. Kammeraat C. Modification of the alkaline picrate assay for creatinine to prevent spuriously elevated values by keto acids. Clin Chem Acta 1978; 84: 119-28.
24. Sahana DA, Shivprakash G, Raghavendra B, et al. Effect of *Eugenia Jambolana* on Plasma Glucose, Insulin Sensititvity and HDL-C Levels: Preliminary results of a randomized clinical trial. J Pharmacy Res 2010; 3: 1268-70.
25. Ansarullah, Bhavna B, Patel V, Ramachandran A.V. *Oreocnide integrifolia* (Gaud.) Miq leaf water extract improves metabolic alterations in high fructose fed insulin resistant and hypertensive rats. Euro J of Integ Med 2010; 2: 79–87.
26. Carl de L, Jerrold MO. Inflammation and Insulin resistance. FEBS Letters 2008; 582: 97-105.
27. Zhiuang XF, Zhao MM, Weng CL, Sun LN. Adipocytokines: A bridge connecting obesity and insulin resistance. Medical Hypothesis 2009; 73: 981-85.
28. Ding EL, Song Y, Malik VS, Liu S. Sex differences of endogenous sex hormones and risk of type 2 diabetes: a systematic review and meta-analysis. JAMA 2006; 295: 288–99.
29. Fukui M, Soh J, Tanaka M, Kitagawa Y, et al. Low serum testosterone concentration in middle-aged men with type 2 diabetes. Endocr J 2007; 54: 871–7.

30. Grossman M, Thomas MC, Panagiotopoulos S, et al. Low testosterone levels are common and associated with insulin resistance in men with diabetes. J Clin Endocrinol Metab 2008; 93: 1834–40.
31. Sharma B, Balomajumder C, Roy P. Hypoglycemic and hypolipidemic effects of flavonoid rich extract from *Eugenia jambolana* seeds on streptozotocin induced diabetic rats. Food Chem Toxicol 2008; 46: 2376-83.
32. Sasa M, Inoue I, Shinoda Y, Takahashi S, et al. Activating effect of momordin, extract of bitter melon (*Momordica Charantia* L.), on the promoter of human PPARdelta. J Atheroscler Thromb 2009; 16: 888-92.
33. McCarty MF. Does bitter melon contain an activator of AMP-activated kinase?. Medical Hypothesis 2004; 63: 340-3.
34. Shibib BA, Khan LA, Rahman R. Hypoglycemic activity of *Coccinia indica ana* and *Momordica charantia* in diabetic rats: depresión of the hepatic gluconeogenic enzymes glucose-6-phosphatase and fructose-1,6-bisphosphatase and elevation of both liver and red-cell shunt enzyme glucose-6-phosphate dehydrogenase. Biochem J 1993; 292: 267-70.
35. Mithieux G, Guignot L, Bordet JC, Wiernsperger N. Intra-hepatic mechanisms underlying the effect of metformin in decreasing basal glucose production in rats fed a high-fat diet. Diabetes 2002; 51: 139-43.
36. Mohamed Farook S and Clement Atlee W. Antidiabetic and hypolipidemic potential of *Tragia Involucrata linn*. in streptozotocin-nicotinamide induced type II diabetic rats. Int J Pharm Pharm Sci 2011; 3: 103-109.
37. MiuraT, Itoh C, Iwamoto N, et al. Hypoglycemic activity of the fruit of the *Momordica charantia* in Type 2 diabetic mice. J Nutr Sci Vitaminol 2001; 47: 340-44.
38. Babu PV, Gokulakrishnan A, Dhandayuthabani R, Ameethkhan D, et al. Protective effect of *Withania somnifera* (Solanaceae) on collagen glycation and cross-linking. Comp Biochem Physiol B Biochem Mol Biol 2007; 147: 308-13.