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PHARMACOLOGICAL EVALUATION OF ANTI-DIABETIC EFFECTS OF COMBINED DOSES OF *MOMORDICA CHARANTIA* AND *STEVIA REBAUDIANA* AGAINST STZ INDUCED DIABETES MODEL

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ABSTRACT: Diabetes mellitus is one of the most prevalent pathogenic conditions which affect around 6% of the world population. Diabetes mellitus is a systemic metabolic disorder, which is characterized by hyperglycemia, hyperlipidemia, hyperaminoacidemia, and hypoinsulinemia. Although, there are different approaches to minimize the ill effects of diabetes and its complications, herbal formulations are favored due to lesser side effects and low cost. Both *Momordica charantia* and *Stevia rebaudiana* are reported to possess the anti-diabetic, however their synergistic or combined effects are unclear. The perception of polyherbalism is given in *Sharangdhar Samhita*: Ayurvedic literature, which dates back to around 1300 AD. Polyherbal treatment improves the therapeutic efficacy and balances the concentrations of individual herbs, thereby reducing adverse events. This research aims to evaluate the anti-diabetic activity of the polyherbal dose of *Momordica charantia* and *Stevia rebaudiana* against their individual doses, the disease control group and the standard drug Glibenclamide. This research also presents the comparative study among the individual and combined doses of *Momordica charantia* and *Stevia rebaudiana* for their anti-diabetic activities in the STZ administered healthy albino rats. In comparison amongst the individual doses, the disease control and the standard drug Glibenclamide, the administration of the combined dose of *Momordica charantia* and *Stevia rebaudiana* show significant decrease (**p<0.005) in the blood glucose, total cholesterol (TC), Triglyceride (TGL), and Low-density lipoprotein (LDL) levels. Though, HDL levels were found to be significantly increased (***p<0.005).

INTRODUCTION: Diabetes is an umbrella term for a number of metabolic disorders and is one of the most common pathogenic conditions affecting the population of both developed and developing countries [1].

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DM is characterized by hyperglycemia which occurs due to abnormalities associated with insulin secretion, insulin action, and insulin uptake by cells [2]. Weight loss, polyurea, polydipsia, polyphagia, blurred vision are few of the most prominent symptoms of hyperglycemia [5]. Complications of long-term hyperglycemia involve nephropathy leading to renal failure, retinopathy with possible loss of vision, autonomic neuropathy causing gastrointestinal, cardiovascular and sexual dysfunction, peripheral neuropathy with the possibility of foot ulcers, amputations, and Charcot joints [2, 6].

Diabetic patients are more susceptible to CVS disorders like Hypertension, Peripheral artery disease (PAD) and atherosclerosis [7].

Based on the type of mechanism underlying the disease pathogenesis, DM falls into two categories. The first category comprises type-1diabetes, in which there is a complete deficit of insulin secretion [8]. Type-1 diabetes is also known as insulin-dependent diabetes, and it accounts for about 5-10% of cases. It occurs due to cellular-mediated autoimmune destruction of the β-cells of the pancreas. The second category comprises type-2 diabetes, in which there is a resistance against insulin action. Type-2 diabetes is also known as non–insulin-dependent diabetes, and it accounts for about 90-95% of cases. In this category individual develop resistance against insulin action and relatively low insulin release [2, 9]. The majority of patients suffering from Type-2 diabetes are obese and obesity is one of the many reasons behind insulin resistance [10]. Type-2 diabetic patients may gradually develop ketoacidosis; generally happens in involvement with the stress of other infections [11]. Type-2 diabetes usually remains asymptomatic due to continuing development of hyperglycemia until any one of the classic symptoms of DM is noticed [12]. According to the WHO, the number of patients suffering from diabetes will get doubled by the year 2025, from the existing figure of about 150 million to 300 million [13].

Momordica charantia is commonly known as bitter gourd, karela and balsam pear. Its fruit is predominantly used in the treatment of diabetes and associated complications by the native populations of Asia, South America, India and East Africa [14]. Additionally, *M. charantia* has significant hypolipidemic activity and thus can be used as an adjuvant along with allopathic treatment of medicine to treat diabetes [15]. *M. charantia* is rich in minerals like potassium, calcium, zinc, magnesium, phosphorus, and iron and is a good source of dietary fiber [16]. Phytochemical screening of *M. charantia* fruits shows the presence of flavonoids, isoflavones, terpenes, anthraquinones, phenols, and glucosinolates, which attributes to its high antioxidant properties. Antidiabetic effects of *M. charantia* are due to the presence of constituents like proteid, triterpene, alkaloid, steroid, inorganic, lipid and phenolic compounds [17, 18].

Stevia rebaudiana is a perennial shrub and is commonly known as sweet leaf. It is a member of the Asteraceae family. It is native to the highlands of Paraguay, where it grows in sandy soils near streams [19]. This medicinal plant is reported to regulate blood sugar, prevent hypertension and treat skin disorders. Additionally, it is known to possess antibacterial and antiviral properties [19, 20].

Extracts of S. *rebaudiana* are primarily used as natural sweeteners in a number of countries [21]. The characteristic sweet taste of *S. rebaudiana* is due to natural sweet active constituents like stevioside and rebaudiosides A, B, C, D, and E; dulcoside A; and steviolbioside. Stevioside has a slightly bitter aftertaste and provides 250-300 times the sweetness intensity of sucrose, and they are noncalorigenic [22, 23]. However, *Stevia* may cause hypoglycemia in diabetic patients since it decreases metabolic processes like glycogenolysis, gluconeogenesis and promotes glucose absorption in duodenum [20].

MATERIALS AND METHODS:

Plant Materials: *Stevia rebaudiana* leaves were collected from fields of Adhar Khera village, near Bakshi Ka Talab, Lucknow (Uttar Pradesh). *Momordica charantia* (Bittergourd) fruits were purchased from a local market in Lucknow (Uttar Pradesh). The plant material was obtained in a sufficient quantity and was authenticated from CIMAP, Lucknow. Every voucher specimen was submitted and preserved in the Department of Pharmacology, Hygia Institute of Pharmaceutical Education and Research, Lucknow. Both *Momordica charantia* fruits and *Stevia rebaudiana* leaves were washed and shade dried (for about 10-15 days) until they became suitable for making powder. Dried plant materials were finely powdered in a grinder and stored in air-tight bottles till further use. The powder obtained was extracted in distilled water using Soxhlet's apparatus.

Animals: In the present study, male albino rats of Wistar strain weighing 200-220 gm were used. Animals were housed in polyvinyl cages of 4 animals per cage were maintained at standard laboratory conditions of temperature (24 ± 2 °C), humidity (40 ± 5%) and 12 h light and dark cycle. Animals had free access to standard pellets and water *ad libitum*.

The experimental protocol was approved by the Institutional Animal Ethics Committee (HIPER/ IAEC/02/17/04) and as per the guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animal (CPCSEA). Throughout the experimental procedure the ethical norms were strictly followed.

Chemicals and Solvents: Glibenclamide tablets and Streptozotocin powder were provided by Cipla Limited (India) and Sigma Aldrich (Germany), respectively.

Streptozotocin-Induced Diabetes Model: Animals were fasted overnight before injecting with streptozotocin. Diabetes mellitus was induced by single intraperitoneal injection of freshly prepared solution of STZ (50 mg/ kg, i.p.) in 0.1 M citrate buffer of pH 4.5 after overnight fasting for 18 h [24, 25]. Rats were administered with oral solution of 5% glucose to combat the early phase of drug-induced hypoglycemia. The glucose levels in blood were analysed after 48 h STZ administration [26]. Fasting glucose level was estimated by using a glucose oxidase-peroxidase reactive strips and a glucometer [27, 28]. Rats with fasting blood glucose levels above 200 mg dL^{-1} and indicating glycosuria were considered diabetic and included in study [29].

Experimental Design: The rats were divided into following six groups containing six rats in each:

Group I (Normal control)	Treated with normal saline for 15 days
Group II (Disease control)	STZ (50 mg/ kg, i.p.) induced diabetic rats treated with normal saline for 15 days
Group III	Diabetic rats treated with standard drug Glibenclamide (10 mg/kg, p.o.) for 15 days.
Group IV	Diabetic rats treated with aqueous extract of *Momordica charantia* (200 mg/kg, p.o.) for 15 days
Group V	Diabetic rats treated with aqueous extract of *Stevia rebaudiana* (200 mg/kg, p.o.) for 15 days
Group VI	Diabetic rats treated with combined aqueous extract of *Momordica charantia* and *Stevia rebaudiana* (400 mg/kg, p.o.) for about 15days.

The fasting glucose levels were determined on days 0, 10 and 15. During the experiment, the rats were weighed daily, and the mean changes in the body weights were calculated. On the 15th day, blood samples were collected from animals (under fasting conditions).

Biochemical Estimation: The serum levels of triglyceride (TGL), total cholesterol (TC) and high-density lipoprotein-cholesterol (HDLC) were determined spectrophotometrically [30], using enzymatic colorimetric assay kits (Randox, Northern Ireland) while low-density lipoprotein cholesterol (LDLC) was calculated [31]. About two milliliters of blood was collected through retro-orbital route from each rat and transferred into an ice-cold centrifuge tubes. The blood samples were centrifuged at 5000 R.P.M for 5 min. The supernatant (serum) collected was assayed for the serum levels of TGL, TC, and HDL-C using the Randox Biochemical kits while LDLC was calculated.

Determination of Blood Glucose Levels in Blood: Fasting blood glucose levels were determined in all experimental rats on the 0, 5, 10 and 15 days to determine the diabetic status. Glucose level was estimated using glucose oxidase peroxidase reactive strips and a glucometer (GOD-POD) [27, 28].

Determination of Triglyceride (TGL) Levels in Blood: Triglyceride (TGL) levels in blood were determined by the method of Treitz [32]. 1 ml of the reagent was added to 10µl each of the sample and standard. The resulting solutions were incubated for 10 min at 20-25 ºC. Later, the absorbance of the sample (A sample) and standard (A standard) was measured against the reagent blank within 30 min.

$$\text{TGL concentration} = A_{sample} / A_{standard} \times 2.29 \text{ mmol/L}$$

Determination of Total Cholesterol (TC) Levels in Blood: The serum level of TC was determined after enzymatic hydrolysis and oxidation of the sample as described by Richmond [33] and Roeschlau *et al.* [34] 1 ml of the reagent was added to 10 µl each of the sample and standard. The resulting solutions were incubated for 10 min at 20-25 °C and the absorbance of the sample (A sample) and standard (A standard) was measured against the reagent blank within 30 min.

$$\text{TC concentration} = A_{sample} / A_{standard} \times 5.17 \text{ mmol/L}$$

Determination of High-Density Lipoprotein-Cholesterol (HDLC) Levels in Blood: Low-density lipoprotein (LDL) and chylomicron

fractions in the sample were precipitated quantitatively by the addition of phosphotungstic acid in the presence of magnesium ions. The resulting mixture was allowed to stand for 10 minutes at room temperature. Later, the mixture was centrifuged for 10 min at 4000 rpm. The supernatant represented the HDLC fraction. The concentration of cholesterol in the HDL fraction, in the supernatant, was determined.

Determination of Low-Density Lipoprotein-Cholesterol (LDLC) Levels in Blood: The concentration (mmol/L) of LDL cholesterol was calculated using Friedewald's equation as stated below [31]:

$$LDLC = TC - (HDLC + TGL/2.2)$$

RESULTS AND DISCUSSION: In this research we used streptozotocin (STZ), a glucosamine–nitrosourea compound derived from *Streptomyces achromogenes* to induce Diabetes in experimental animals. STZ is a cytotoxic nitrosourea compound and it damages pancreatic β-cells by its alkylating property. It is supposed that STZ is taken up by GLUT2 glucose transporter; causes DNA alkylation and eventually results in β cell death [35]. The damage of β-cells and initiation of the hyperglycemia is linked with inflammatory infiltrates like lymphocytes in the pancreatic islets [36]. One of the several pathogenic mechanisms underlying the pathogenesis of Diabetes mellitus is the autoimmune destruction of the β-cells of the pancreas resulting in insulin deficiency, which leads to the disturbance in carbohydrate, fat and protein metabolism [3]. Insulin deficiency may occur due to insufficient insulin secretion and/or diminished tissue responses to insulin [4]. Since ancient times, a number of plants and plant extracts are used to alleviate the ailments of human race. The aim of the present research is to study the effect of individual and combined dose of *Stevia rebaudiana* and *Momordica charantia* (Bittergourd) on streptozotocin (STZ)-induced diabetic rats by measuring blood glucose and alternations in the lipid profile.

TABLE 1: EFFECT OF INDIVIDUAL AND COMBINED DOSES OF *MOMORDICA CHARANTIA (MC)* AND *STEVIA REBAUDIANA (S)* ON THE BLOOD GLUCOSE LEVELS OF RATS IN STZ MODEL

S. no.	Groups	Blood glucose levels: 0-day (mg/dl)	Blood glucose levels: 5-days (mg/dl)	Blood glucose levels: 10-days (mg/dl)	Blood glucose levels: 15-days (mg/dl)
1	Normal Control	87.6±10.19	86.00±11.52	85.03±12.13	84.98±12.13
2	Disease control	207.66±11.76$^{\#}$	209.51±11.39$^{\#}$	211.77±12.29$^{\#}$	213.75±13.41$^{\#}$
3	Standard	121.10±37.57**	115.73±25.25	112.00±22.74	106.69±14.30
4	Treatment-1(Mc)	94.15±34.40^{NS}	93.85±9.39^{NS}	91.16±8.28^{NS}	91.00±8.14^{NS}
5	Treatment-2(S)	94.35±9.03	93.79±9.48	93.34±8.24	92.00±8.25
6	Combination (Mc+S)	91.89±9.18**	89.16±10.39**	89.70±10.52**	87.16±11.28**

Values are given as Mean ± SEM of experimental animals (n=6); NS: Non significance. $^{\#}$P<0.05: Represents statistical significance against Normal Control, **P<0.05: Represents statistical significance against Disease Control.

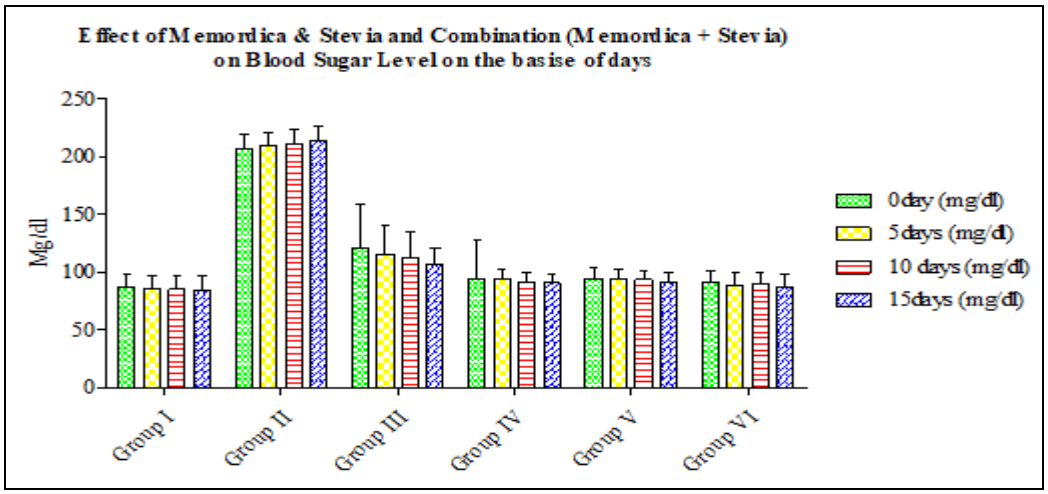

FIG. 1: EFFECT OF COMBINATION (MC AND S) ON BLOOD GLUCOSE LEVEL IN RATS. Values are given as mean ± SEM of experimental animals (n=6); (#p<0.005 *vs.* Control), (**p<0.005 *vs.* Disease control). Data was analysed by One-way Anova followed by tukey's test.

Assessment of Blood Glucose Levels: Blood glucose levels gradually increase from day 0, 5, 10 and 15 in the disease control group (#p<0.005) as compared to control group. As compared to the disease control and the standard group, the blood glucose levels significantly decrease (**p<0.005) on the administration of both individual (200 mg/kg, p.o.) and combined (400 mg/kg, p.o) doses of *M. charantia* and *S. rebaudiana.*

Assessment of TC, TGL, LDL and HDL Levels in Blood: As compared to the disease control and the standard, blood levels of total cholesterol (TC), Triglyceride (TGL), and Low-density lipoprotein (LDL) were found to be significantly reduced (# p<0.005) on the administration of combined (400 mg/kg, p.o) doses of *Momordica charantia* and *Stevia rebaudiana.*

TABLE 2: EFFECT OF INDIVIDUAL AND COMBINED DOSES OF *MOMORDICA CHARANTIA (MC)* AND *STEVIA REBAUDIANA (S)* ON THE BLOOD TOTAL CHOLESTEROL, TRIGLYCERIDE, LDL AND HDL-CHOLESTEROL OF RATS IN STZ MODEL

S. no.	Groups	Total Cholesterol (mg/dl)	Triglyceride Level (mg/dl)	LDL-Cholesterol (mg/dl)	HDL-Cholesterol (mg/dl)
1	Normal Control	102.62±5.41	89.70±5.30	42.03±4.13	42.98±5.13
2	Disease Control	160.15±8.76$^{\#}$	140.51±8.39$^{\#}$	110.77±3.29$^{\#}$	20.75±3.41$^{\#}$
3	Standard	109.10±7.57**	105.73±9.25	45.25±4.16	40.59±4.30
4	Treatment-1 (Mc)	134.95±8.40^{NS}	122.15±9.19	60.85±4.28	35.32±3.14
5	Treatment-2 (S)	127.95±9.35 ^{NS}	110.79±8.48	55.34±3.24	37.80±4.25
6	Combination(Mc+S)	104.89±9.18**	98.16±6.39	44.70±4.24	41.64±4.38

Values are given as Mean ± SEM of experimental animals (n=6); NS: Non significance. $^{\#}$P < 0.05: Represents statistical significance against Normal Control, **P< 0.05: Represents statistical significance against Disease Control

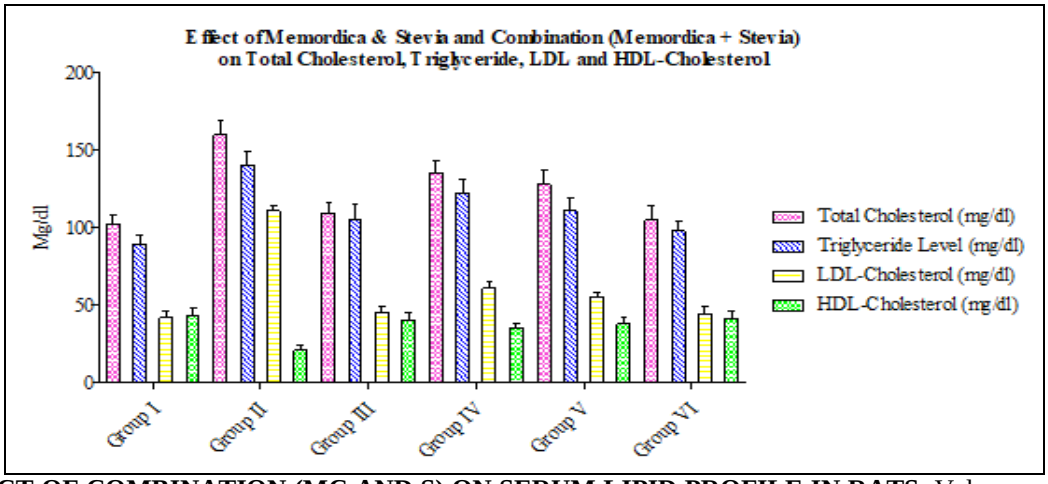

FIG. 2: EFFECT OF COMBINATION (MC AND S) ON SERUM LIPID PROFILE IN RATS. Values are given as mean ± SEM of experimental animals (n=6); (#p<0.005 *vs.* control), (**p<0.005 *vs.* disease control). Data were analyzed by One-way Anova followed by Tukey's test

CONCLUSION: The data obtained from the current research clearly indicates that the current combination therapy involving *Momordica charantia* and *Stevia rebaudiana* can be used as a possible therapeutic approach that may block the disease pathogenesis and alleviate pathology in *Diabetes Milletus*.

The current combination therapy synergistically decreases the blood glucose, Total cholesterol (TC), Triglyceride (TGL), and Low-density lipoprotein (LDL) levels in the treatment groups. However, further studies are helpful in developing and understanding the more evident pathways which may enhance the synergistic action and therapeutic efficacy associated with the current combination therapy.

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REFERENCES:

1. Hameed I, Masoodi SR, Mir SA, Nabi M, Ghazanfar K and Ganai BA: Type-2 diabetes mellitus: From a metabolic disorder to an inflammatory condition. World J Diabetes 2015; 6(4): 598-12.
2. Ormazabal V, Nair S, Elfeky O, Aguayo C, Salomon C and Zuniga FA: Association between insulin resistance and the development of cardiovascular disease. Cardiovasc Diabetol 2018; 17(1): 122.
3. Akbarzadeh A, Norouzian D and Mehrabi MR: Induction of diabetes by Streptozotocin in rats. Indian J Clin Biochem 2007; 22(2): 60-4.
4. Deeds MC, Anderson JM, Armstrong AS, Gastineau DA, Hiddinga HJ, Jahangir A, Eberhardt NL and Kudva YC: Single dose streptozotocin-induced diabetes: considerations for study design in islet transplantation models. Lab Anim 2011; 45(3): 131-40.
5. Rajeswari G and Rajagopalan V: Evaluation of anti-diabetic effects of *C. zizanioides* Linn root extracts in streptozotocin induced diabeteic Wistar rats. Journal of Scientific and Innovative Research 2013; 2(3): 555-74.
6. Edwards JL, Vincent AM, Cheng HT and Feldman EL: Diabetic neuropathy: mechanisms to management. Pharmacol Ther 2008; 120(1): 1-34.
7. Thiruvoipati T, Kielhorn CE and Armstrong EJ: Peripheral artery disease in patients with diabetes: Epidemiology, mechanisms, and outcomes. World J Diabetes 2015; 6(7): 961-9.
8. Wu J and Yan LJ: Streptozotocin-induced type-1 diabetes in rodents as a model for studying mitochondrial mechanisms of diabetic β cell glucotoxicity. Diabetes Metab Syndr Obes 2015; 8(1): 181-88.
9. Parveen S, Khan M, Odigwe CC, Qureshi F and Iroegbu N: Latent Autoimmune Diabetes of Adult Masquerading as Type 2 Diabetes Mellitus. Journal of Medical Cases 2015; 6(7): 327-29.
10. Daousi C, Casson IF, Gill GV, MacFarlane IA, Wilding JP and Pinkney JH: Prevalence of obesity in type-2 diabetes in secondary care: association with cardiovascular risk factors. Postgrad Med J 2006; 82(966): 280-84.
11. Hallett A and Modi A: Developments in the management of diabetic ketoacidosis in adults: implications for anaesthetists. BJA Education 2016; 16 (1): 8-14.
12. Pearl GL and Jeffrey JB: The pathophysiology of hyperglycemia in older adults: Clinical Considerations. Diabetes Care 2017; 40(4): 444-52.
13. Alvarenga KF, Duarte JL, da Silva DPC, Agostinbo-Pesse RS, Negrato CA and Costa OA: Cognitive P300 potential in subjects with diabetes mellitus. Brazilian Journal of Otorhinolaryngology 2005; 71(2): 202-07.
14. Cefalu WT, Ye J and Wang ZQ: Efficacy of dietary supplementation with botanicals on carbohydrate metabolism in humans. Endocr Metab Immune Disord Drug Targets 2008; 8 (2): 78-81.
15. Joseph B and Jini D: Antidiabetic effects of Momordica charantia (bitter melon) and its medicinal potency. Asian Pac J Trop Dis 2013; 3(2): 93-102.
16. Snee LS, Nerurkar VR, Dooley DA, Efird JT, Shovic AC and Nerurkar PV: Strategies to improve palatability and increase consumption intentions for *Momordica charantia* (bitter melon): A vegetable commonly used for diabetes management. Nutr J 2011; 10(1): 78.
17. Saeed MK, Shahzadi I, Ahmad I, Ahmad R, Shahzad K and Ashraf M: Nutritional analysis and antioxidant activity of bitter gourd (*Momordica charantia*) from Pakistan. Pharmacologyonline 2010; 1(1): 252-60.
18. Budrat P and Shotipruk A: Extraction of phenolic compounds from fruits of bitter melon (*M. charantia*) with subcritical water extraction and antioxidant activities of these extracts. Chiang Mai J Sci 2008; 35(1): 123-30.
19. Kujur RS, Singh V, Ram M, Yadava HN, Singh KK, Kumari S and Roy BK: Antidiabetic activity and phytochemical screening of crude extract of *Stevia rebaudiana* in alloxan-induced diabetic rats. Pharmacognosy Res 2010; 2(4): 258-63.
20. Assaei R, Mokarram P and Dastghaib S: Hypoglycemic effect of aquatic extract of Stevia in pancreas of diabetic rats: PPARγ-dependent Regulation or Antioxidant Potential. Avicenna J Med Biotechnol 2016; 8(2): 65-74.
21. Kolb N, Herrera JL, Ferreyra DJ, Uliana RF: Analysis of sweet diterpene glycosides from *Stevia rebaudiana*: Improved HPLC method. Journal of Agricultural and Food Chemistry 2001; 49 (10): 4538-41.
22. Goyal SK, Samsher and Goyal RK: Stevia (*Stevia rebaudiana*) a bio-sweetener: a review. Int J Food Sci Nutr 2010; 61(1): 1-10.
23. Thiyagarajan M and Venkatachalam P: Large scale *in-vitro* propagation of *Stevia rebaudiana* (bert) for commercial application: Pharmaceutically important and antidiabetic medicinal herb. Ind Crops Prod 2012; 37(1): 111-17.
24. Gajdosik A, Gajdosikova A, Stefek M, Navarova J and Hozova R: Streptozotocin-induced experimental diabetes in male Wistar rats. Gen Physiol Biop 1999; 18(1): 54-62.
25. Rizk SM, El-Maraghy SA and Nassar NN: A novel role for SIRT-1 in L-arginine protection against STZ induced myocardial fibrosis in rats. PLoS One 2014; 9(12): e114560.
26. Gotama TL, Husni A and Ustadi: Antidiabetic activity of *Sargassum hystrix* extracts in streptozotocin-induced diabetic rats. Prev Nutr Food Sci 2018; 23(3): 189-95.
27. Tonyushkina K and Nichols JH: Glucose meters: a review of technical challenges to obtaining accurate results. J Diabetes Sci Technol 2009; 3(4): 971-80.
28. Kermani SK, Khatony A, Jalali R, Rezaei M and Abdi A: Accuracy and Precision of Measured Blood Sugar Values by Three Glucometers Compared to the Standard Technique. J Clin Diagn Res 2017; 11(4): OC05-OC08.
29. Qinna NA and Badwan AA: Impact of streptozotocin on altering normal glucose homeostasis during insulin testing in diabetic rats compared to normoglycemic rats. Drug Des Devel Ther 2015; 9(1): 2515-25.
30. Penumarthy S, Penmetsa GS and Mannem S: Assessment of serum levels of triglycerides, total cholesterol, high-density lipoprotein cholesterol, and low-density lipoprotein cholesterol in periodontitis patients. J Indian Soc Periodontol 2013; 17(1): 30-5.
31. Friedewald WT, Levy RI and Fredrickson DS: Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem 1972: 18(6): 499-02.
32. Tietz NW: Clinical Guide to Laboratory Tests, W.B. Saunders Company, Philadelphia, USA. Edition 2, 1990: 554-56.
33. Richmond N: Preparation and properties of a cholesterol oxidase from Nocardia sp. and its application to the enzymatic assay of total cholesterol in serum. Clin Chem 1973; 19(12): 1350-56.
34. Roeschlau P, Bernt E and Gruber WL: Kinetics of the cholesterol-sulfuric acid reaction. A fast kientic method for serum cholesterol. Clin Che Clin Bio 1974; 12(1): 403-08.
35. Dufrane D, van Steenberghe M, Guiot Y, Goebbels RM, Saliez A and Gianello P: Streptozotocin-induced diabetes

in large animals (pigs, primates): role of GLUT2 transporter and β-cell plasticity. Transplant 2006; 81(1): 36-45.
36. Paik SG, Fleischer N and Shin SI: Insulin-dependent diabetes mellitus induced by subdiabetogenic doses of streptozotocin: obligatory role of cell-mediated autoimmune processes. Proc Natl Acad Sci USA 1980; 77(10): 6129-33.

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