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ANTIDIABETIC AND LIPID LOWERING EXTENUATING IMPACT OF *GLYCINE MAX* LEAVES (SOYABEAN) IN TYPE II DIABETES MELLITUS SUBJECTS

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ABSTRACT: Diabetes mellitus is possibly the world's largest growing metabolic disease. Currently, a challenge is to identify such healthy foods that remain in the realms of obscurity and to establish them as functional foods to prevent the progression of metabolic complications. *Glycine* max is indigenous to India and is used extensively in the traditional system of medicine to treat diabetes, reduces coronary heart disease, lowers cholesterol level and other myriad ailments. Therefore, the objective of the study was undertaken to evaluate the effect of *Glycine max* leaves powder on type II diabetes mellitus subjects. The present study was conducted on type II diabetes mellitus subjects of the middle-income group which were selected from the main campus of Banasthali Vidyapith, Newai Rajasthan and divided into two groups, *i.e*., experimental (A) and control (B) groups (Age group: 40-60 years). *Glycine max* 10g leaves powder was incorporated in biscuits and administered to the experimental group daily for 60 days. Biochemical evaluations of blood sample of subjects were done. The present study indicates that *Glycine max* leaves powder showed a significant effect on blood glucose and lipid parameters. There is a significant decrease in pre and postprandial blood glucose level, glycosylated Hb, total cholesterol, total triglycerides, LDL-C, VLDL-C. There is no significant change in HDL-C may be due to the short period of supplementation. *Glycine max* purports to alleviate the symptoms of diabetes naturally with no adverse effects on health and low cost or inexpensive than the other hypoglycemic drugs.

INTRODUCTION: Diabetes mellitus (DM) is a chronic metabolic disorder that represents a serious public health concern. It is characterized by defective insulin secretion or deficiencies in the action of insulin. Patients with type II diabetes frequently have some metabolic abnormalities including insulin resistance, hypertension, dyslipidemia, hyperuricemia, and coagulopathy. The underlying mechanisms that lead to the clustering of these abnormalities are not well understood.

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Genetic factors are implicated, but environmental factors such as diet are also important [1, 2, 3]. The prevalence of diabetes mellitus has now reached epidemic proportions in both developed and developing countries, affecting more than 366 million people suffer from DM and the number is expected to rise to 552 million by 2030.[4]

The International Diabetes Federation (IDF) estimates the total number of diabetic subjects to be around 40.9 million in India and this is further set to rise to 69.9 million by the year 2025.[5] Hyperglycemia, the common characteristic of both type 1 diabetes mellitus (IDDM) and type 2 diabetes mellitus (NIDDM) have the potential to cause serious complications due to its insidious and chronic nature [6]. Diabetes mellitus associated dyslipidemia is a major factor responsible for the development of macrovascular complications [7].

Many indigenous Indian medicinal plants are useful to successfully manage diabetes. One of the great advantages of medicinal plants is that these are readily available and have very low side effects [8]. The soybean (*Glycine max*) belongs to Leguminosae [9] is indigenous to East Asia, widely grown for its edible bean [10]. Soy protein products can replace animal-based food possessing complete proteins; however, it contains more fat, especially saturated fat without requiring major adjustments elsewhere in the diet [11]. Soybean contains approximately 40-45% protein and 18-22% oil [12] and is a rich source of vitamins and minerals. *Glycine max* seeds are used as an ethnomedicine for treating diabetes by tribal people of Tamil Nadu [13].

The role of soy in the prevention of CVD, particularly LDL cholesterol-lowering effects, has been the subject of numerous controlled clinical studies [14]. In 2006, study [15] reported findings from a 1-year trial in which 66 individuals who adhered well to the portfolio diet (31.8% of participants) experienced reduced serum LDL cholesterol levels by 29.7%. The beneficial effects which have been documented include decreased low-density lipoprotein (LDL) concentrations, triglycerides, lipoprotein, C-reactive protein, homocysteine, oxidized LDL, blood pressure and increased high-density lipoprotein (HDL) concentrations [16, 17, 18].

Epidemiological studies in Japanese women suggested that consumption of soy products has a protective effect against menopausal symptoms [19]. Studies in Asia revealed that women in Shanghai, China, who ate the sumptuous amount of soy foods, were one third less likely to experience a fracture than Chinese women who consumed a lower amount of soy [20]. Several nutritional intervention studies in animals and humans indicated that consumption of soy protein reduces body weight and fat mass in addition to lowering plasma cholesterol and triglycerides. In obese humans, dietary soy protein also reduces body weight and body fat mass in addition to reducing plasma lipids [21].

Hence in the present study, an attempt has been done to investigate the hypoglycemic and hypolipidemic effect of *Glycine max* leaves powder in type II diabetes mellitus.

MATERIALS AND METHODS:

Collection of Plant Material: The leaves of *Glycine max* was collected from the botanical garden of Banasthali Vidyapith Rajasthan, India.

Chemicals: The chemicals and solvents used in the present study were purchased from Sigma Chemical Co. (Saint Louis, MO, USA), HiMedia Labs. (L.B.S Marg Mumbai) and Merck Chemicals in Mumbai, India. All the chemicals and solvents were of analytical grade.

Experimental Design:

Participants: Twenty patients with type II diabetes (aged 40-60 years) including both male as well as female were recruited from the Banasthali village area and all the diabetic subjects were randomly divided into two groups, *i.e*., Experimental group (A) and control group (B) with ten subjects each. Exclusion criteria included the following: Fasting blood glucose levels, as well as glycosuria, were assessed to confirm the diabetic state; inability to consume the provided biscuit supplementation, body mass index (BMI).

Procedures: Baseline data (urine collections, lipids profile, hemoglobin (HbA1c) were collected while patients were on their usual diets. Then, subjects were randomized to the biscuit supplementation. Group A was supplemented with 10g *Glycine max* leaves powder in the form of biscuits, and Group B was supplemented with biscuits but without incorporation of *Glycine max* leaves powder for 60 days respectively. After 60 days, biochemical evaluations of subjects were done. Approximately 5 ml of fasting blood sample was collected from each subject on day 0 and day 60 and was used for further evaluation. All the initial tests were repeated at days 19 to 21. Blood glucose was measured with elegance glucometer (CT-X10, Convergent Technologies, Germany) at 0 days and 60 days after daily administration of leaves powder of *Glycine max* incorporated in biscuits. Subjects were also invited to return 2 months after completing the study for repeat testing to determine whether there was a sustained effect of the supplementation.

Blood Glucose and Lipid profile: On day 60, blood was collected by glucometer. The values were expressed as mg/dl of blood with glucometer

was assayed by the method [22]. Blood glucose level was estimated by GOD/ POD enzymatic method [23]. Glycosylated hemoglobin (HbA1c) was estimated by ion exchange resin method [24]. Total cholesterol was estimated by CHOD-PAP method [25]. HDL-cholesterol was estimated by PEG-CHOD-PAP method [26]. Total triglyceride was estimated by Mc Gowan method [27]. LDL and VLDL were estimated by Friedewald equation [28].

Statistical Analysis: All the data were expressed as mean ± SD. Statistical analysis was carried using Student's t-test to analyze the significance between the groups.

RESULTS AND DISCUSSION: This study was carried out to observe the hypoglycemic and hypolipidemic effect of *Glycine max* leaves powder in type II diabetes mellitus subjects.

Effect of *Glycine max* Leaves Powder on Fasting Blood Glucose and Post Prandial Blood Glucose Level in Diabetic Subjects: The result shows a significant decrease at both level ($p \leq 0.01$) and ($p \leq 0.05$) in the mean fasting blood glucose and postprandial blood glucose level. The calculated value of t is greater than the tabulated value; the hypothesis is accepted. Hence, the supplement is useful for diabetic subjects.

TABLE 1: EFFECT OF *GLYCINE MAX* LEAVES POWDER ON FASTING BLOOD GLUCOSE LEVEL AND POSTPRANDIAL BLOOD GLUCOSE LEVEL IN DIABETIC SUBJECTS

	Group	Pre intervention (Mean ± SD) (mg/dl)	Post intervention (Mean ± SD) (mg/dl)	Mean (Difference between pre and post intervention)	t-test
Fasting blood glucose level	A (Experimental)	151 ± 8.45	144.9 ± 8.27	-6.1	9.76**
	B (Control)	179.0 ± 5.90	180.9 ± 6.41	+1.88	0.833*
Post prandial glucose level	A (Experimental)	263.33 ± 19.47	254.1 ± 18.09	-9.23	7.06**
	B (Control)	252.8 ± 0.76	253.6 ± 0.45	+0.76	0.10*

**Significant level ($p \leq 0.01$) and ($p \leq 0.05$), *Non-Significant

Effect of *Glycine max* Leaves Powder on Glycosylated Hb Level in Diabetic Subjects: The result shows a significant decrease at both level ($p \leq 0.01$) and ($p \leq 0.05$) in the mean glycosylated Hb level. The calculated value of t is greater than the tabulated value; the hypothesis is accepted. Hence, the supplement is useful for diabetic subjects.

TABLE 2: EFFECT OF *GLYCINE MAX* LEAVES POWDER ON GLYCOSYLATED HB LEVEL IN DIABETIC SUBJECTS

Group	Pre-intervention (Mean ± SD) (mg/dl)	Post-intervention (Mean ± SD) (mg/dl)	Mean (Difference between pre and post intervention)	Paired t-test
A	11.21 ± 0.52	9.94 ± 0.57	-1.28	7.89**
B	6.93 ± 1.07	6.93 ± 0.98	0	0.94*

**Significant level ($p \leq 0.01$) and ($p \leq 0.05$), *Non-Significant

Effect of *Glycine max* Leaves Powder on Total Cholesterol and Triglyceride Level in Diabetic Subjects: The result shows a significant decrease at both level ($p \leq 0.01$) and ($p \leq 0.05$) in the mean total cholesterol and triglyceride level. The calculated value of t is greater than the tabulated value; the hypothesis is accepted. Hence, the supplement is useful for diabetic subjects.

TABLE 3: EFFECT OF *GLYCINE MAX* LEAVES POWDER ON TOTAL CHOLESTEROL LEVEL AND TOTAL TRIGLYCERIDE LEVEL IN DIABETIC SUBJECTS

	Group	Pre intervention (Mean ± SD) (mg/dl)	Post intervention (Mean ± SD) (mg/dl)	Mean (Difference between pre and post intervention)	Paired t-test
Total cholesterol level	A	225.83 ± 8.23	221.20 ± 8.72	-4.63	7.39**
	B	219.9 ± 15.56	221.6 ± 9.52	+1.7	0.10*
Total triglyceride level	A	211.99 ± 20.38	202.89 ± 21.87	-8.8	6.53**
	B	149.1 ± 10.08	151.3 ± 5.81	+2.2	0.10*

**Significant level ($p \leq 0.01$) and ($p \leq 0.05$), *Non-Significant

Effect of *Glycine max* Leaves Powder on Total HDL-Cholesterol, Total LDL-Cholesterol and Total VLDL-Cholesterol Level in Diabetic Subjects: The result shows no change at both level ($p \leq 0.01$) and ($p \leq 0.05$) in the mean total HDL cholesterol level.

Hence, the supplement didn't get affected and stabilize the HDL-C in diabetic subjects. The result shows a significant decrease at both levels ($p \leq 0.01$) and ($p \leq 0.05$) in the mean total LDL and VLDL cholesterol level. The calculated value of t is greater than the tabulated value; the hypothesis is accepted. Hence, the supplement is useful for diabetic subjects.

TABLE 4: EFFECT OF *GLYCINE MAX* LEAVES POWDER ON TOTAL HDL-CHOLESTEROL, TOTAL LDL-CHOLESTEROL AND TOTAL VLDL-CHOLESTEROL LEVEL IN DIABETIC SUBJECTS

	Group	Pre intervention (Mean ± SD) (mg/dl)	Post intervention (Mean ± SD) (mg/dl)	Mean (Difference between pre and post intervention)	Paired t-test
Total	A	54.35 ± 1.24	55.43 ± 1.10	+0.95	2.68**
HDL level	B	46.7 ± 5.84	46.1 ± 4.68	-0.57	0.11*
Total	A	129.06 ± 9.88	125.26 ± 10.64	-3.8	6.37**
LDL level	B	144.3 ± 9.62	146.2 ± 5.30	+3.2	0.09*
Total	A	42.39 ± 4.07	40.63 ± 4.48	-1.76	6.81**
VLDL level	B	26.8 ± 5.22	32.9 ± 4.78	+6.1	0.09*

**Significant level ($p \leq 0.01$) and ($p \leq 0.05$), *Non-Significant

Glycine max possesses many properties, and this plant may procure at a large scale for providing an herbal alternative to many diseases. Herbal drugs are prescribed widely because of their effectiveness, fewer side effects, and relatively low cost. Therefore, investigation on such agents from traditional medicinal plants has become more important. Keeping given traditional uses, the powder of *Glycine max* leaves was analyzed for its antidiabetic and hypolipidemic activities. Significant reduction of blood glucose levels was observed in diabetic subjects treated with *Glycine max* leaves powder incorporated in biscuits.

Similarly, the extract of *Glycine max* at (200 and 500 mg/kg) exhibited a dose-dependent significant anti-hyperglycemic activity on the 21st day of post-treatment. Treatment with extract for 3 weeks showed a significant reduction in levels of total cholesterol, triglycerides, low-density lipoprotein, and very low-density lipoprotein. The study concluded that seed extracts of *Glycine max* possess significant antidiabetic activity as well as antihyperlipidemic activity [29]. The results were similar to the study of a comparable hypoglycemic effect was evidenced that fasting blood glucose and doses of 200 and 400 mg/kg b.w produced at par reduction of in blood glucose [30]. There is a significant decrease in total cholesterol and serum triglycerides respectively.

There is no significant decrease seen in HDL-cholesterol. The results were also similar to the study that intake of soybean foods containing soybean protein up to 36 g/day or above and isoflavones up to 52 mg/day is assumed to decrease total serum cholesterol (3.77% - 9.3%), LDL cholesterol (3% - 12.9%) triacylglycerol (10.5%) and increased HDL cholesterol (2.4%) [31, 32, 33, 34]. However, they have used higher doses. The lipid-lowering effect of leaves powder of *Glycine max* in our study has a useful effect which avoids the complication of diabetes.

CONCLUSION: From this study, we can conclude that *Glycine max* leaves powder supplementation decreased fasting and postprandial blood glucose level, glycosylated hemoglobin, total cholesterol, total triglyceride, LDL-C, VLDL-C and there was no significant effect on HDL-C parameter due to a short period of supplementation. *Glycine max* has a beneficial role in diabetes having no ill effects on human health, easily available. Hence, it may be a safe and better alternative available over other agents in diabetes-associated dyslipidemia and can be further used as a dietary supplement.

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