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Triphala improves glucose homeostasis by alleviating atherogenic lipids and oxidative stress in human Type 2 diabetes mellitus

Research Article

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Abstract

Aims: 'Triphala' constituting equal parts of three medicinal dried plant fruits *Emblica Officinalis* Gaertn., *Terminalia chebula* Retz. and *Terminalia bellerica* Gaertn. is an antioxidant rich Ayurvedic formulation. The present study assessed therapeutic as well as protective effects of Triphala on human subjects with Type 2 diabetes mellitus (T2DM) and Impaired glucose tolerance (IGT).

Materials and methods: Triphala at a dose of 5 gms BD was administered to two cohorts viz., IGT, N= 20 and T2DM, N=30 consecutively for a period of 12 months. The therapeutic efficacy was assessed quarterly by monitoring blood glucose and lipid levels; the protective effect by monitoring antioxidants level quarterly and DNA damage annually. Toxicity if any, to liver and kidney due to long term administration was assessed quarterly in both cohorts.

Results: Continuous 'Triphala' therapy for 12 months significantly reduced blood glucose ($p \leq 0.001$) and lipid levels ($p \leq 0.05$) in both the cohorts. Triphala resisted oxidative stress generated during the course of hyperglycemia by significantly increasing the activity of super oxide dismutase and Catalase ($p \leq 0.001$) and the level of reduced glutathione ($p \leq 0.001$). Protective effect on DNA was accessed through significant reduction in the comet tail length ($p \leq 0.001$).

Conclusions: 'Triphala' ameliorated not only the oxidative stress but also normalized glucose and lipid homeostasis in subjects with impaired glucose and T2DM.

Keywords: Type 2 diabetes, impaired glucose tolerance, Lipid metabolism, Glucose metabolism, Oxidative stress, Antioxidants, Ayurvedic formulations

Introduction

Type 2 Diabetes mellitus (T2DM) is characterised as a metabolic disorder as it alters both glucose and lipid homeostasis (1, 2). Globally, diabetic population outnumber owing to high degree of genetic predisposition and physical inactivity, characterised by high abdominal adiposity, body fat percentage and level of insulin resistance (3, 4). The most common alterations in lipid profile in T2DM involve an elevation in plasma triglyceride (TG) and Low density lipoprotein cholesterol (LDL-C) concentrations, and low levels of High density lipoprotein cholesterol (HDL-C) (5, 6, 7). Hypertriglyceridemia contributes significantly to the increased risk for premature cardiovascular disease in T2DM (8). Perturbed glucose and lipid homeostasis are manifested as oxidative stress in the insulin dependent as well as insulin independent tissues (9). Development of impaired glucose tolerance (IGT) is the outcome of the cumulative effect of all above mentioned disparities and worsened into the form of fully developed T2DM.

Diabetes Control and Complications Trial (DCCT) and the United Kingdom Prospective Diabetes Study (UKPDS) have demonstrated improved outcomes with early detection and tight control of blood glucose (10, 11). Diabetes Epidemiology: Collaborative analysis of Diagnostic criteria in Europe (DECODE) stated that subjects with IGT have an increased risk for developing T2DM and cardiovascular complications and therefore form an important high-risk group for actions aimed at preventing T2DM (12).

According to WHO, traditional herbal remedies may provide valuable leads for the development of alternative drugs and strategies for T2DM and its related complications (13). Herbs prove good antidiabetic agents as they not only improve glucose and lipid

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metabolism by stimulating insulin release; attenuating the absorption of carbohydrates from the gut but also improve antioxidant status. 'Triphala' is the most commonly used Indian Ayurvedic herbal formulation of the "Rasayana" group, consisting equal parts of three medicinal dried plant fruits *Emblica Officinalis* Gaertn., *Terminalia chebula* Retz. and *Terminalia bellerica* Gaertn. (14). Triphala is reported to exhibit a variety of biological activities, such as anti-cancer, anti-mutagenic, anti-viral, anti-oxidant and free radical scavenging activities and is reported to be reno- and hepato-protective (15, 16). Triphala and its individual components have been shown to prevent hyperglycemia and diabetic cataract (17, 18). The present study evaluated the therapeutic potential of 'Triphala' in human subjects with IGT and T2DM.

Materials and Methods

Study design and Participants

This was a unicentre, randomized and prospective study comparing two different modalities, IGT and T2DM subjects. The study was conducted in diabetes clinic run by School of Studies in Biochemistry, Jiwaji University, Gwalior, India, on every sunday under the supervision of an Ayurvedic physician. About 500 T2DM and IGT subjects are registered and regularly attend the camp for monthly check up and follow up. The study was approved by the human ethical committee board of Jiwaji University Gwalior. The study protocol was approved by AYUSH and Institutional Human Ethics Committee.

Selection criteria for the study included the following for the people with T2DM and IGT; age >40 years, not on any antihyperglycemic or antioxidant therapy, not taking any lipid lowering therapy. A total of 50 subjects attending the diabetes clinic were selected and registered for the study after monitoring their glycemic status and were categorised to one of the two modalities viz., IGT subjects (FBG: 110-126 mg/dl and PPBG: ≥140 mg/dl to ≤200 mg/dl, N= 20) and T2DM (FBG: ≥126mg/dl and PBG: ≥200mg/dl, N= 30) with poor glycemic control based on the criteria for the diagnosis of T2DM and IGT by the WHO. The mean age of the subjects was 54.3±1.75 years in T2DM group and 55.1±2.02 years in IGT group. The duration of diabetes was 4.4±0.58 years for T2DM subjects and 3.8±0.66 years for IGT subjects. All the subjects were informed of the objectives of the study prior to registration and were asked to give a written informed consent. Other demographic and clinical characteristics of the participating subjects are given in Table 1. Subjects suffering from any chronic or acute inflammatory illness were excluded from the study.

Preparation of Triphala composition

Fresh mature fruits of *Emblica Officinalis* Gaertn. were purchased from local market in the Gwalior and shade dried. Whole dried fruit of *Terminalia chebula* Retz. and *Terminalia bellerica* Gaertn. were purchased from the local market in the Gwalior. The dried fruits were then separately milled to fine powder; the powder of individual components was further mixed in the equal proportion to get the Triphala composition.

Administration of 'Triphala'

The registered T2DM and IGT subjects were enrolled under Triphala therapy at a dose of 5 gms BD before meal (30 minutes prior) every day with normal water for a period of 12 months. Triphala powder was packaged in the airtight sealed plastic bags and each packet contained 35 gms Triphala powder (5 gms BD for 7 days) for the first three months. After three months of weekly follow up, sincere and regular patients were then continued for the next nine months of study and each packet contained 150 gms of Triphala powder (5 gms BD for 30 days) till the study end; medicine did not change during the study period. The participants were provided regular counselling on causes and management of diabetes besides anti diabetic ayurvedic medicines. None of the study subjects were on antioxidant supplementation or lipid lowering or anti-diabetic drugs while taking Triphala. The subjects were advocated to maintain their moderate physical activity and to avoid carbohydrate rich diet during the study period. The subjects were followed up at regular monthly intervals for up to 12 months. The therapeutic efficacy of 'Triphala' was evaluated by monitoring glycemic control and plasma lipid profile at specific intervals during the course of study.

Anthropometric data collection and measurement methods

Details of ethnic origin and age were directly ascertained from the subjects; waist (cm), at the level of the umbilicus with the patient standing and breathing normally, while weight (kg) with weighing balance and height (m), with scale and metal rule, were measured (in light clothing, without shoes). After 10 minutes rest, systolic (first phase) and diastolic (fifth phase) blood pressures (sBP, dBP) were taken on the dominant arm in a sitting position, using a standard mercury gauge sphygmomanometer.

Collection of blood samples

Approximately 2.5 ml of venous blood was collected in the morning after 12 hours fast from each subject under study once in the beginning of the study and at monthly intervals during the course of study period. Fasting blood samples were transferred to sterilized centrifuge tubes containing sodium EDTA/ sodium fluoride (for glucose) and allowed to stand for 10 minutes at room temperature and then centrifuged (5000 rpm x 10 min x 4^{o}c) to get clear plasma. Plasma samples were stored at -20^{o}C for later analysis. After

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removal of plasma and buffy coat from the whole blood; the red blood cells were suspended in 2 ml normal saline and centrifuged (3000 rpm x 10 min x 4°C) to get cell pellet. The cell pellet was washed thrice with normal saline to get packed cell volume (PCV). Haemolysate was prepared by mixing 1.9 ml of cold distilled water to 0.1 ml of PCV suspension and stored at 4˚C.

Laboratory methods

Blood glucose concentrations in fasting and postprandial samples were determined by the glucose oxidase and peroxidase method using a commercial kit (Erba diagnostics glucose kit, Mannheim Germany) in the fresh blood samples at monthly intervals. **Glycosylated Hemoglobin** was measured from fresh blood at 3 monthly intervals by ion exchange resin method. The key **lipid profile markers** viz. total serum cholesterol (TC) (Cholesterol oxidase- peroxidase method), serum triglyceride (TG) (Glycerol 3 peroxidase method), Serum High Density Lipoprotein Cholesterol (HDL-C) (Polyethylene Glycol Precipitation method), Low Density Lipoprotein Cholesterol (LDL-C) (Freidewald's Formula) and Very Low Density Cholesterol (VLDL-C) (Freidewald's Formula) were assayed using commercial kits (Ecoline Merck NJ USA) from stored plasma at 4 monthly intervals. Analysis of **liver function markers** viz., Serum aspartate aminotransferase (AST), Alanine amino transferase (ALT) by Modified International federation of clinical chemistry method and serum Bilirubin by Modified Jendrassik and Grof's method at 4 monthly intervals from stored plasma. **Kidney function markers** viz., Urea by Glutamate Dehydrogenase kinetic method, uric acid by Uricase Method and creatinine by Alkaline Picrate Method were analyzed in serum samples at 4 monthly intervals from stored plasma. **Oxidative stress markers** viz., Superoxide dismutase activity and Catalase activity were analysed at 4 monthly intervals from the hemolysate (19, 20). Lipid peroxidation and reduced glutathione were estimated by from fresh whole blood. Protein was estimated by the method of Lowry et al (21, 22).

Statistical analysis

The sample size of at least 20 participants per group provided adequate power to detect a difference. Statistical analysis was performed using one-way ANOVA and comparison of the data was done by Student-Newman-Keuls Method with all pair-wise multiple comparison procedures. The data were expressed as Mean ± SE. The level of significance was set at $p \leq 0.05$.

Results

Baseline characteristics

Table 1 shows the baseline characteristics of the 50 subjects enrolled in the study which includes 30 T2DM and 20 IGT subjects. FBG and PPBG were significantly higher in T2DM compared to IGT subjects ($p \leq 0.05$). In addition systolic blood pressure was significantly higher in T2DM subjects ($p \leq 0.05$).

Table 1: Baseline characteristic features of subjects with IGT and T2DM

Features	IGT	T2DM
Number	20	30
Age (years)	55.1±2.02	54.3±1.75
Sex (male/female)	16/4	28/2
Weight in (Kg)	67.8±2.03	71.2±2.09
Body mass index BMI (kg/m^2)	24.8±0.46	25.6±0.55
Fasting blood glucose (mg/dl)	124.5 ±0.6	172.3±6.6
PP blood glucose (mg/dl)	168.9 ±4.18	272.3±11.4
Systolic blood pressure (mm Hg)	137.6±3.21	141.6±3.17
Diastolic blood pressure (mm Hg)	80.4±2.58	81.6±0.99
Duration of disease (years)	3.8±0.66	4.4±0.58

Results: mean values (± S.E.M); IGT: Impaired glucose tolerance; T2DM: Type 2 diabetes mellitus.

Glucose metabolism

Blood glucose, HbA1c variables for month 0, 4, 8 and 12 for both the treatment groups are shown in Table 2. Individual group analysis revealed that the baseline FBG and PPBG value were significantly higher in the T2DM compared to the IGT subjects ($p \leq 0.001$). Triphala therapy significantly reduced the glucose levels in the circulation however in T2DM subjects the reduction in PPBG levels 29.0% ($p \leq 0.001$) was more pronounced compared to the FBG 20.9% ($p \leq 0.001$) by 12^{th} month; however, 50% and 57% of this decrease occurred by 4^{th} month in both PPBG and FBG respectively. Similarly, in subjects with IGT the FBG and PPBG levels were significantly reduced by 23.5% ($p \leq 0.001$) and 22.3% ($p \leq 0.001$) by 12^{th} month of Triphala therapy however 57% and 64% of this reduction occurred from 4^{th} month onwards in FBG and PPBG respectively. Glycemic level did not affect the blood glucose lowering potential of Triphala as drawn out from the significant reductions in FBG and PPBG in both T2DM and IGT groups from 4^{th} month onwards. GHb levels showed 22.7% ($p \leq 0.001$) and 18.1% ($p \leq 0.001$) reduction in T2DM and IGT subjects; however, 41% and 43% of this decrease occurred from 4^{th} month onwards indicating that short term administration of Triphala showed good glycemic control in T2DM and IGT subjects respectively.

Table 2: Effect of 12 months of Triphala therapy on blood glucose and HbA1c in subjects with T2DM and IGT.

Variable(s)	Intervals (months)	IGT (n=20)	T2DM (n=30)
FBG (mg/dl)	0	124.5 ±0.6	172.3±6.6
	4	107.9±4.21[b]	151.7±6.5[c]
	8	107.2±4.22[b]	148.0±5.3[c]
	12	95.2±1.79[a]	136.2±5.2[a]
P.P BG (mg/dl)	0	168.9 ±4.18	272.3±11.4
	4	144.5±5.49[b]	232.8±12.0[c]
	8	137.9±5.98[a]	214.5±8.2[c]
	12	131.2±4.95[a]	193.3±8.0[a]
HbA1c (%)	0	6.5±0.22	9.6±0.35
	4	6.0±0.22	8.7±0.35[c]
	8	5.6±0.20[b]	8.3±0.39[c]
	12	5.4±0.21[a]	7.4±0.33[a]

Values are mean ±SE; [a]p£0·001, [b]p£0·01, [c]p£0·05 are significantly different from baseline value (0 month) by one way ANOVA.

Lipid metabolism

Analysis of individual group reveals glycemic control was associated with a significant improvement in plasma lipid profile. Fasting plasma TG, TC, LDL-C, levels were significantly higher in T2DM patients than in IGT group and consumption of Triphala led to significant and time dependent improvement (Table 3). By 12th month atherogenic lipids such as TC and LDL-C were significantly reduced by 23.1% ($p\leq0.001$) and 27.4% ($p\leq0.001$) respectively in T2DM subjects; however 28.3% and 27.4% of this reduction was observed by 4th month, indicative of effectiveness of long term administration of Triphala in the management atherogenic lipids in subjects with T2DM. By 12th month subjects with IGT showed 21% ($p\leq0.001$) and 36.2% ($p\leq0.001$) reduction in TC and LDL-C levels; however, 54% and 50% decrease of this value was observed in 4th month, indicative of effectiveness of short term administration of Triphala in subjects with IGT. HDL-C is significantly increased by 21.1% ($p\leq0.05$) and 20.2% ($p\leq0.05$) in T2DM and IGT group; however, 32% and 31% of this increase was observed in 4th month. The total population of T2DM and IGT on Triphala displayed a marked reduction in the mean total plasma TG by 25.9% ($p\leq0.05$) and 22.1% ($p\leq0.05$) in 12 months; however, 29.4% and 32% of this decrease was observed in 4th month. The data indicates that long term Triphala therapy was beneficial for both IGT and T2DM.

Table 3: Effect of Triphala therapy on Lipid profile in subjects with T2DM and IGT.

Variable(s)	Intervals (months)	IGT (n=20)	T2DM (n=30)
TC (mg/dl)	0	174.7 ±4.65	191.8±6.84
	4	155.0±5.34[b]	179.3±6.60
	8	149.7±4.25[b]	160.4±5.27[a]
	12	138.0±5.41[a]	147.4±5.35[a]
TG (mg/dl)	0	141.0 ±9.76	158.9±10.71
	4	130.9±9.63	146.7±10.68
	8	114.6±9.45[c]	133.3±10.94[c]
	12	109.7±9.00[c]	117.6±10.18[c]
HDL-C (mg/dl)	0	36.7 ±2.59	35.8±2.24
	4	39.0±2.76	37.2±2.36
	8	41.8±3.02	38.5±2.09
	12	46.1±2.70[c]	44.6±2.0[c]
LDL-C (mg/dl)	0	109.7 ±5.59	125.2±6.67
	4	89.7±6.53[c]	112.7±6.25
	8	84.9±5.34[b]	95.2±4.81[a]
	12	69.9±5.79[a]	79.6±4.85[a]
VLDL-C (mg/dl)	0	28.2 ±1.95	31.7±2.14
	4	26.2±1.92	29.3±2.13
	8	22.9±1.89[c]	26.6±2.18[c]
	12	21.9±1.80[c]	23.5±2.03[c]

Values are mean ±SE; [a]p£0·001, [b]p£ 0·01, [c]p£0·05 are significantly different from baseline value (0 month) by one way ANOVA.

Alleviation of Oxidative stress

The activity of antioxidant enzymes viz., SOD and CAT were significantly increased by 31.3% ($p\leq0.001$) and 34.2% ($p\leq0.001$) in T2DM subject; however, 30.4% and 45.6% of this increase was observed in 4th month of Triphala therapy. IGT subjects showed 33.6% ($p\leq0.001$) and 37% ($p\leq0.001$) increase in the activity of SOD and CAT in 12 months however 31% and 40% of this increase was observed in 4th month respectively. By 12th month the level of GSH was increased significantly in both T2M and IGT subjects by 34.5% ($p\leq0.001$) and 33.7% ($p\leq0.001$) respectively, by 4th month 33.7% of this increase was observed in both T2DM and IGT subjects. TBARS levels were significantly reduced by 25.5% ($p\leq0.001$) and 30% ($p\leq0.01$) in T2DM and IGT subjects respectively by 12th month, however 26% and 28% of this decrease was observed in 4th month of Triphala therapy. Triphala therapy retrieved the oxidative damage of DNA quantitatively as evident from the significant reductions in the comet tail length by

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21.1% (p£0·005) in T2DM and 26% (p£0·001) in IGT subjects respectively. Hence the results indicate that short term administration of an antioxidant rich natural therapy like Triphala can significantly improve the antioxidant status.

Table 4: Effect of Triphala therapy on oxidative stress markers markers.

Variable(s)	Intervals (months)	IGT (n=20)	T2DM (n=30)
Superoxide Dismutase (SOD) (Unit/min/ mg of protein)	0	0.83±0.04	0.76 ±0.02
	4	0.91±0.04	0.84±0.03[c]
	8	1.10±0.05[a]	0.97±0.05[c]
	12	1.25±0.05[a]	1.11±0.06[a]
Catalase (CAT) (µmole/min/ mg protein)	0	6.7±0.32	6.5 ±0.19
	4	7.7±0.34[c]	7.5±0.26[c]
	8	9.2±0.45[b]	8.2±0.39[a]
	12	10.6±0.26[a]	9.8±0.36[a]
Reduced glutathione (GSH) (mg/ ml)	0	1.9±0.13	1.8 ±0.11
	4	2.2±0.18[c]	2.0±0.15
	8	2.6±0.21[c]	2.5±0.15[b]
	12	3.1±0.1[a]	2.8±0.12[a]
Thio-barbeturic acid reactive substance (TBARS) (moles of MDA / ml blood)	0	368.5±22.02	451.5 ±18.04
	4	337.0±23.11[c]	421.3±16.48
	8	301.1±21.19[c]	371.1±16.27[b]
	12	257.7±16.99[b]	336.1±16.27[a]

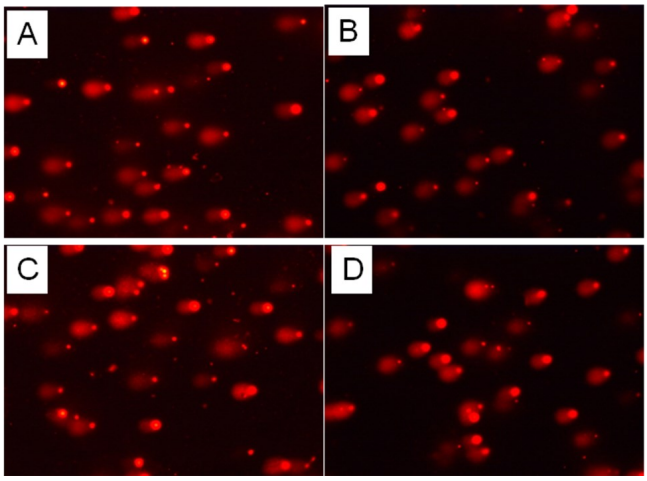
Values are mean ±SE; [a]p£0·001, [b]p£ 0·01, [c]p£0·05 are significantly different from baseline value (0 month) by one way ANOVA.

Table 5: Effect of Triphala therapy on oxidative DNA damage after 12 months

Variable(s)	Intervals	IGT (n=20)	T2DM (n=30)
Comet tail length (arbitrary unit)	0 month	4.6±1.6	5.2 ±1.1
	12 months	3.4±1.02[b]	4.1±1.07[a]

Values are mean ±SE; [a]p£0·001, [b]p£ 0·01, [c]p£0·05 are significantly different from baseline value (0 month) by one way ANOVA.

Figure 1: Assessment of DNA damage through alkaline comet assay in Triphala administered subjects. Ethedium bromide stained representative pseudo colored images are shown for IGT (A) and T2DM (C) respectively before Triphala Regimen. Intense red color represents intact DNA, while blurred tail like structure shows increasing level of DNA fragmentation. Images (B) are (D) are captured under identical magnification (10x) to show the relative recovery after Triphala therapy respectively.



Antitoxic effect on Liver and kidney

The liver function markers viz., Bilirubin, SGOT and SGPT were significantly reduced by 22.2% (p≤0.001), 22.1% (p≤0.05) and 29% (p≤0.01) respectively in T2DM subjects, similarly IGT subjects showed 25% (p≤0.001), 24% (p≤0.05) and 28% (p≤0.01) reduction in the following liver function markers respectively.

Kidney function markers viz., creatinine, urea and uric acid were significantly reduced by 27% (p≤0.001), 29% (p≤0.001) and 24% (p≤0.001) in T2DM subjects. Similarly, IGT subjects showed significant reduction of 24% (p≤0.01), 25% (p≤0.001) and 26% (p≤0.001) respectively in 12 months of Triphala therapy. Results indicated antitoxic effect of long term administration of Triphala on liver and kidney.

Table 6: Effect of Triphala therapy on Liver and kidney function markers.

Variable(s)	Intervals (months)	IGT (n=20)	T2DM (n=30)
AST (U/I)	0	27.2±1.83	26.6 ±1.46
	4	25.0±1.69	22.9±1.73[c]
	8	23.1±1.90	24.1±1.66[c]
	12	20.7±1.76[c]	20.7±1.42[c]
ALT (U/I)	0	26.4±1.63	28.4 ±1.42
	4	24.1±1.45[c]	23.4±1.60[c]
	8	22.5±1.51[c]	22.8±1.60[c]
	12	18.9±1.33[b]	20.1±1.10[b]

Bilirubin (mg/dl)	0	0.8±0.03	0.96±0.03
	4	0.75±0.03	0.81±0.02[a]
	8	0.66±0.03[c]	0.76±0.03[a]
	12	0.6±0.03[a]	0.75±0.03[a]
Urea (mg/dl)	0	33.3±1.12	35.2 ±1.15
	4	30.0±1.46	30.7±1.01[b]
	8	28.3±1.46[c]	29.9±1.07[b]
	12	24.9±1.06[a]	25.0±1.02[a]
Uric acid (mg/dl)	0	5.3±0.21	5.4 ±0.17
	4	4.6±0.17[c]	4.8±0.16[c]
	8	4.2±0.17[a]	4.4±0.14[b]
	12	3.9±0.16[a]	4.1±0.16[a]
Createnine (mg/dl)	0	1.04±0.06	1.09 ±0.04
	4	0.96±0.05	0.95±0.04[c]
	8	0.93±0.04	0.91±0.04[c]
	12	0.79±0.04[b]	0.79±0.04[a]

Values are mean ±SE; [a]p £0·001, [b]p £ 0·01, [c]p £0·05 are significantly different from baseline value (0 month) by one way ANOVA.

Discussion

Diabetes mellitus is the World's largest growing metabolic disorder, goes un-noticed in the form of prediabetics (IGT), presenting all the aetiologies of T2DM, thus forms a major risk group aimed at preventing T2DM. Traditional plant medicines are being used in place of costlier modern medicines throughout the world for better management of T2DM and IGT (23). Thus in the present study antioxidant rich ayurvedic formulation Triphala was validated for its antihyperglycemic, antihyperlipidemic and antioxidant potential in human T2DM and IGT subjects.

Oral administration of Triphala to humans T2DM and IGT subjects for 12 months resulted in a significant reduction in both FBG and PPBG levels however; the reduction in PPBG was more pronounced compared to FBG in IGT groups. The improvement in glycemic levels of IGT group due to Triphala can help control the progression of T2DM. Treatment with Triphala significantly decreased the levels glycosylated haemoglobin. The proportion of haemoglobin that is glycosylated is known to increase substantially in conditions of sustained hyperglycaemia (24, 25). The significant reduction in glycemic level with Triphala could be due to the optimization of activities of carbohydrate metabolizing enzymes, increase in carbohydrate absorption time and stimulation of insulin release from the pancreatic beta cells (26, 27). Glucose lowering effects of Triphala have been reported earlier in STZ induced diabetic rats (27, 28, 29).

The American Heart Association emphasizes the role of lipid abnormalities in T2DM (30, 31). In the present study dyslipiemia was recorded in both T2DM and IGT though the increase in triglycerides (TG) and low-density lipoprotein cholesterol (LDL-C) levels and decrease high-density lipoprotein cholesterol (HDL-C) was more in T2DM. We observed the percentage improvement in lipid profile owing to Triphala was statistically significant in both IGT and T2DM subjects. Both treatment groups with reduction in levels of plasma TG and TC displayed a significant increase in the levels of the HDL fraction on Triphala therapy. The decreased serum HDL-C was reversed towards normalization after Triphala therapy. The beneficial effect of Triphala on HDL–C is well recognised and likely to be clinically relevant. The progressive shift of LDL profile towards normal with decrease in fasting TG and TC levels observed in T2DM resulted perhaps from a specific decrease in plasma levels of glucose fractions by Triphala. The blood glucose and lipid lowering activity of Triphala demonstrated in IGT subjects in this study may represent a novel and important preventive approach in delaying the onset of diabetes mellitus and CVD; however the long term administration of Triphala proved effective in addressing dyslipidemia. The blood lipid lowering effects of Triphala are attributed to *Terminalia chebula* Retz. (32, 33)

Results of the present study indicated that Triphala besides antihyperglycemic, antihyperlipidemic possesses a very good antioxidant property. Triphala administration attenuated the diabetes induced depletion of antioxidants. Triphala not only significantly increased the activity of antioxidant enzymes viz., SOD and CAT but also increased the levels of reduced glutathione in both the treatment groups. The observed improvement in the level of these antioxidants after the administration of Triphala in hyperglycemia may be due to the direct reaction of gallic acid, ascorbic acid and flavonoids in Triphala with the free radicals generated and converting the reactive oxygen free radicals to non-reactive products (34, 35). Lipid peroxidation is regarded as one of the basic mechanism of tissue damage caused by free radicals. Triphala administration significantly reduced the levels of LPO in both T2DM and IGT groups. This scrutiny reveals that Triphala is able to quench the LPO chain and is capable to shield the membrane from free radical caused injuries. Triphala therapy significantly protected the DNA from oxidative damage in both T2DM and IGT subjects. This present study thus indicates the antioxidant potency of the drug.

To conclude, the results of the present study have empirically indicated that Triphala has protective role in IGT and was effective in the treatment of T2DM. A consistent decrease in the blood glucose level by Triphala in diabetic patients and parallel improvement in lipid profile makes it a good choice as an alternative antidiabetic drug which is non habit forming and cheap as alternatives are needed due to side effects of several

allopathic drugs and development of resistance to currently used.

There are clearly some limitations of this study. First, this is an ancillary study that is dependent on the main study for the study design. Second the number of patients in each group is small. Though the study was powered for antihyperglycemic potential of the drug under observation, a very important parameter i.e. measurement of insulin release is lacking.

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Conflict of interest

The authors declare that there is no conflict of interest associated with this manuscript.

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