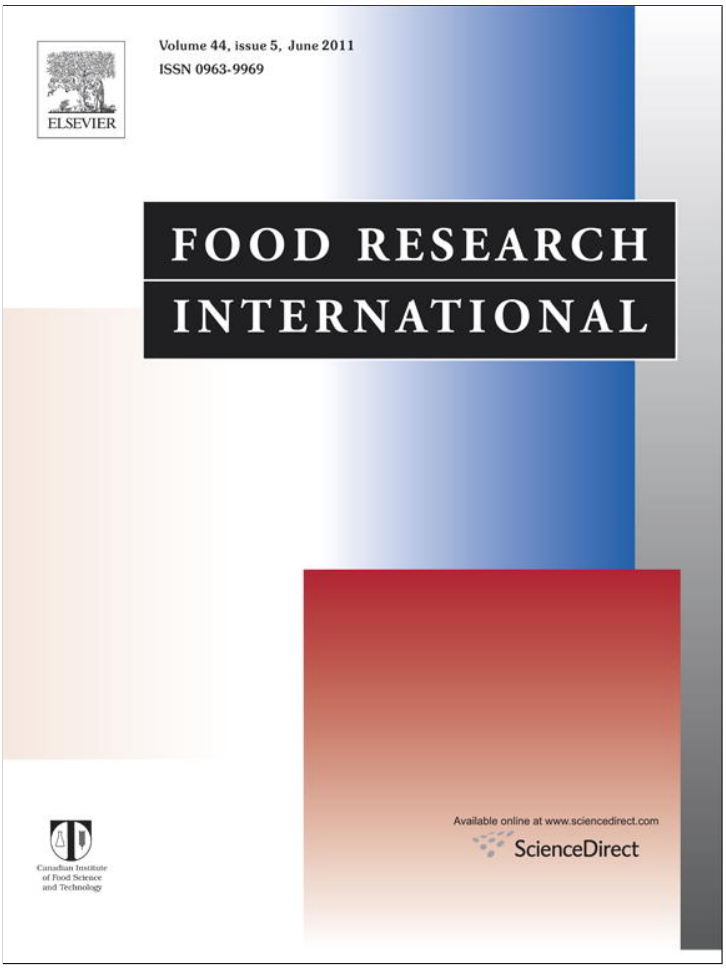
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Effects of *Dichrostachys glomerata* spice on cardiovascular diseases risk factors in normoglycemic and type 2 diabetic obese volunteers

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ABSTRACT

Previous *in vivo* and *in vitro* experimental studies have shown *Dichrostachys glomerata* (*DG*), a spice used in western Cameroon, to have potential antioxidant and hypoglycemic properties. The purpose of the present study was to evaluate the effects of orally administered *DG* on various cardiovascular disease risk factors in obese normoglycemic and obese type 2 diabetic human subjects. The study was an 8 week randomized, double-blind, placebo-controlled design with obese and obese/diabetic participants (20 males, 72 females, ages 25–65). The subjects were randomly divided into four groups: 2 normoglycemic obese groups (active; placebo) and two type 2 diabetic obese groups (active; placebo). Capsules containing the active (400 mg *DG*) or placebo formulation were administered 30–60 min before lunch and dinner throughout the study period. A total of 7 anthropometric and hemodynamic as well as 7 biochemical measurements were taken at the beginning of the study and after 4 and 8 weeks of treatment. All diabetic patients maintained their prior lifestyle intervention and dietary control for the duration of the study. Compared to the two placebo groups, the two active groups showed statistically significant differences on all 14 variables between Weeks 0 and 8. These included body weight, BMI, waist and hip circumference, body fat, blood pressure, blood cholesterol, triglycerides, glucose, and glycosylated hemoglobulin. The results confirm the hypothesis that *DG* appears to reduce cardiovascular disease risk factors in obese normoglycemic and obese type 2 diabetic human subjects.

1. Introduction

Obesity, an ever-increasing phenomenon in both industrialized and developing nations, is considered a significant risk factor for many major morbidities such as coronary heart disease, hypertension, cancer, respiratory complications, osteoarthritis, stroke, and adult-onset (type 2) diabetes (Popkin, 2001, 2008).

Type 2 diabetes is considered a part of the metabolic syndrome, which comprises a cluster of interrelated common clinical disorders including impaired glucose tolerance, insulin resistance, hyperglycemia, dyslipidemia, hypertension, obesity or abdominal fat accumulation, prothrombotic and proinflammatory states (Reilly & Rader, 2003; Tenenbaum, Fisman, & Motro, 2003). Hyperglycemia and dyslipidemia progress to atherogenesis, with consequent macrovascular and microvascular complications (Reilly & Rader, 2003; Tenenbaum et al., 2003). Thus people with diabetes are at an increased risk of cardiovascular, peripheral vascular and cerebrovascular diseases (Alberti & Zimmet, 1998). Hypertension is common in diabetes, with an increased risk of diabetic nephropathy, as well as microvascular and macrovascular complications (Bakris, Sowers, Epstein, & Williams, 2000). Patients with type 2 diabetes are characterized by resistance to weight loss due to overactivity of the endocannabinoid system (Matias et al., 2006; Wing, Marcus, Epstein, & Salata, 1987) and to increased cardiovascular risk (NCEP-ATPIII, 2002; Haffner, Lehto, Ronnemaa, Pyorala, & Laakso, 1998), with obesity considered an additional and independent risk factor.

Risk of cardiovascular disease is 200% higher in the diabetic than non-diabetic population (Grundy et al., 2002), and coronary heart disease is the leading cause of death among diabetic patients (Wilson et al., 1998; Gu, Cowie, & Harris, 1998). Western diets are high in fat and carbohydrates and tend to promote obesity. Increased intake of high caloric (energy and fat) food promotes body fat storage and greater body weight and adiposity in humans (Bray et al., 2002). Weight reduction is associated with favorable changes in cardiovascular risk factors (Vidal, 2002). It is not only the cornerstone in the prevention of type 2 diabetes, but it also improves glycemic control and reduces other risk factors associated with diabetes/metabolic syndrome. Even modest weight loss following lifestyle changes or medical treatment has beneficial effects on several cardiovascular risk factors (Fujioka, 2002).

Although diet and lifestyle changes remain the cornerstone of therapy for obesity and type 2 diabetes (Van Gaal, 1998), weight loss is often minimal and long-term success is extremely disappointing. Over the last decade, obesity and diabetes research has focused on the exploration of new biochemical pathways and pharmacological intervention possibilities, but despite a wide variety of dietary manipulations (e.g., overall caloric restriction, specific fat restriction,

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very low calorie diets, etc.), the long-term maintenance of clinically significant weight loss (defined as a loss of 5–10% of initial body weight) remains unsatisfactory. In contrast, many medicinal foods have been successfully used since ancient times to counteract diabetes and its associated complications (Bailey & Day, 1989; Campos-Vega, Loarca-Pina, & Ooma, 2010). Due to their availability, effectiveness and safety, medicinal foods are widely prescribed even when their biologically active compounds are unknown (Valiathan, 1998).

D. glomerata (*DG*) (Forssk.) is a deciduous tree of western Cameroon whose fruits and seeds are edible. The fruits are dry dehiscent constricted pods commonly used as spices in "Nah po," a traditional soup eaten with taro (Tchiégang & Mbougueng, 2005). The hypotensive property of the plant was reported more than four decades ago (Roth & Keller, 1963). Recent studies in our laboratory have shown the fruit to have *in vitro* and *in vivo* antioxidant activity, along with LDL oxidation inhibiting property (Kuate, Etoundi, Soukontoua, Ngondi, & Oben, 2010). It has also been shown to reduce fasting serum glucose levels and lower glycosylated hemoglobin in experimental diabetic rats (Kuate, 2010). The present study was undertaken to investigate the effects of *DG* spice on cardiovascular risk factors in obese individuals with and without type 2 diabetes.

2. Materials and methods

2.1. Test materials: D. glomerata (DG)

All test materials were bought from the local market in Yaoundé, Cameroon. Dried pods of *DG* were ground and encapsulated in individual packets. The identical-looking placebo and active formulation capsules each contained 400 mg of maize-based powder consisting of either maltodextrin or *DG* formulation. Since the capsules were identical in size, shape, and appearance, neither the researchers nor participants knew which treatment was given.

2.2. Study design/intervention

The study was a randomized, double-blind, placebo-controlled design lasting 8 weeks. A total of 46 obese-normoglycemic and 46 obese-diabetic participants completed the study. The volunteers were randomly divided into four equal groups: two normoglycemic obese (OB) groups and two type 2 obese-diabetic (OB/DB) groups. Subjects in the two active formulation groups took one 400 mg *DG* capsule 30–60 min before lunch and dinner throughout the study period, while subjects in the two control groups took a placebo capsule.

2.3. Study population

Participants for the study were recruited from the city of Yaoundé and its surrounding metro region via radio and print advertisement. Eligible subjects included males and non-pregnant/non-lactacting females aged 25–65 years, with a BMI>30 kg/m^2. Diagnosis of the type 2 obese/diabetic subjects were based on clinical characteristics; i.e., the presence of obesity (BMI >30 kg/m^2), no history of ketosis or strong family history of diabetes. Moreover, all patients with type 2 diabetes met the World Health Organization diagnostic criteria of either an abnormal oral glucose tolerance test, or two abnormal fasting blood glucose (>7.0 mmol/L-126 mg/dL) or the American Diabetic Association criteria of high HbA1c (>7%). All obese/diabetic patients maintained their prior lifestyle interventions and dietary controls throughout the study.

2.4. Exclusion criteria

Exclusion criteria included impaired kidney function, cardiac problems, serious hypertension (systolic and diastolic blood pressure blood pressure above 180 mm Hg, and 110 mm Hg, respectively), need for daily insulin management, and enrollment in another clinical study within the past 6 months. Also excluded (following a physical examination) were volunteers who had been on cholesterol-lowering, inflammation-reducing and/or other medications (e.g., steroids) that interfered with healing 30 days prior to enrolling in the study. Also excluded were participants with an active infection, a systemic disease such as HIV/AIDS, active hepatitis or clinical signs of active malignancy within the past 5 years, medical conditions known to affect serum lipids, a history of drug or alcohol abuse, and/or involvement in intense exercise programs.

2.5. Approval; informed consent

The study was approved by the Cameroon National Ethics Board. The purpose, nature, and potential risks of the study were explained to all participants, who gave their written informed consent before participation. The study was done in full compliance with the Declaration of Helsinki (1983 version).

2.6. Anthropometric measurements

The various anthropometric measurements were recorded at baseline and at biweekly follow-up visits for 8 weeks. Height was measured with a Harpended™ stadiometer, which measures the length of curved line staffage to the nearest 0.5 cm. Body weight and percentage body fat were assessed using a Tanita^{TM} BC-418 Segmental Body Composition Analyzer/Scale that uses bio-electrical impedance analysis for body composition analysis. BMI was calculated as the ratio of weight to the height in metres square. The participants (12-hour fasted) were asked to wear light clothing when measurements were taken. Waist and hip measures were obtained with a soft non-stretchable plastic tape. Waist circumference was measured to the nearest 0.1 cm on the narrowest and the widest parts of the trunk; hip circumference was obtained at the widest point of the hip. In an effort to ensure intra-individual consistency, the participants were measured at approximately the same time of day each visit by the same technician.

2.7. Blood pressure

As with the anthropometric measurements, blood pressure was recorded at baseline and at biweekly follow-up visits for 8 weeks. Blood pressure was measured on the left arm after a 10-minute rest. Repeated readings were taken at 5-minute intervals, for 3 sitting measurements, and the average was recorded.

2.8. Sample collection

After a 12-hour overnight fast, blood samples were collected into heparinized tubes at baseline and after 4 and 8 weeks of treatment. The plasma obtained from each blood sample (5 ml of blood) was split into multiple 500 μl aliquots and stored at −20 °C until it was needed for the measurement of lipid profile and fasting blood glucose levels. Whole blood and packed cells (RBCs) were used for total hemoglobin and glycosylated hemoglobin assays.

2.9. Analytical methods

The Trinder glucose activity test (1969), which determines glucose in the blood using glucose oxidase with an alternative oxygen receptor, was adopted for this study. Plasma total cholesterol was assayed by the cholesterol oxidase method (Richmond, 1973), while triglycerides, as well as serum glucose levels, were assayed following the method described by Bucolo and David (1973). HDL cholesterol was determined using a heparin manganese precipitation of Apo B-containing lipoproteins (Bachorik et al., 1976), and LDL cholesterol

was calculated using the Friedewald formula (Friedewald, Levy, & Fredrickson, 1972). Hemoglobin and glycosylated hemoglobin (HbA_1C) were estimated, respectively, by the methods of Drabkin and Austin (1932), and Sudhakar, Nayak, and Pattabiraman (1982) as modified by Bannon (1982).

2.10. Statistical analysis

The data were summarized (mean, standard error of mean) for Week 0 (Initial), Week 4 and Week 8 (final) and for the intra-group variation, and analyzed by SPSS 16.0 for Windows (SPSS Inc., Chicago, Illinois, USA). Because the repeated measurements on each participant were correlated (covariance) a mixed model approach—a flexible tool for analyzing repeated and longitudinal treatments—was used to assess variation between and within patients. Statistical analyses were performed on three monthly data sets (Weeks 0, 4, 8) to determine differences between groups, as well as the differences in variable changes over time between groups. The interaction between time and intervention was tested at the 0.05 level of significance. If the interaction was significant, comparisons were made between the treated and placebo groups for each month. Paired *t* tests were used to test for within-treatment changes from initial values, whereas between-treatment changes were tested using 1-way analysis of covariance (ANCOVA), adjusted for baseline values, with the initial value as a covariate.

3. Results

3.1. Baseline characteristics

The baseline (T0) anthropometric, hemodynamic, and metabolic characteristics of the four study groups are listed in Tables 1–8. Plasma triglyceride, total cholesterol, plasma glucose and the artherogenicity index (TC/HDL-C) were higher, and HDL cholesterol was lower, in the two diabetic groups (Tables 5 and 6). The baseline experimental variables in subjects randomized to treatment with *DG* or placebo within each diagnostic group were essentially not significantly different.

3.2. Anthropomorphic characteristics; blood pressure

Tables 1 and 2 show the changes in the various anthropometric variables (body weight, BMI, waist and hip circumference, and percent body) fat over the 8-week trial period. Compared to the placebo groups, both the obese normoglycemic (OB) and obese type 2 diabetic group (OB/DB) showed a significant weight loss ($p<0.001$) after 8 weeks of treatment with *DG*. The changes in BMI, waist and hip circumferences and body fat paralleled the loss in weight. Blood pressure decreased significantly compared with placebo groups ($p<0.001$) in association with weight loss in both treated subgroups; the decrease appeared unrelated to diagnostic category or amount of weight loss (Tables 3 and 4).

3.3. Blood sample (serological) characteristics; glycosylated hemoglobin

As shown in Tables 5 and 6, there were significant within-group variations in lipid profile from baseline to 8 weeks of *DG* treatment in triglyceride, total cholesterol (TC), HDL cholesterol (HDL-C), and LDL cholesterol (LDL-C) concentrations. These variations were significant compared with the placebo groups ($p<0.001$). Due to the combined changes in both HDL-C and TC concentration, the TC/HDL-C ratio decreased more after weight loss in both treated groups. There was also a significant reduction of blood glucose in both groups; the reduction was greater in the OB/DB participants after 8 weeks of treatment ($p<0.001$). Glycosylated hemoglobin levels were lower in obese non diabetic patients (Table 7) compared to obese type 2 diabetic patients (Table 8). As shown in Table 8, glycosylated hemoglobin was reduced by DG treatment in the OB/DB group compared with placebo ($p<0.001$).

With regard to lipid profile (Tables 5 and 6), the strong hypolipidemic effects of *DG* in the two test groups (OB and OB/DB) over the 8-week period suggest its potential protection against cardiovascular diseases through a reduction in Total Cholesterol [TC]: mean = −43.09 mg/dL (−23.73%) and −72.6 mg/dL (−31.97%); LDL-C: 50.22 mg/dL (−39.27%) and −84.77 mg/dL (−49.56%); Triglyceride [TAG]: mean = −55.43 mg/dL (−41.31%) and −79.09 mg/dL (−56.10%); and an increase in HDL-C: mean = 25.58 (126.06%) and 39.57 (155.88%). An increase in HDL-C alone has been shown to impart possible health benefits in overweight and obese people and reduce the risk for cardiovascular disease (Combaret et al., 2004). The increase in HDL-C, coupled with a decrease in concentrations of LDL-C and triglycerides, could lead to a lowering of atherogenicity. This, in turn, might result in a decrease in the potential incidence of coronary heart disease as highlighted by the reduction of the TC/HDL-C ratio: mean = −3.64 (51.92%) and −6.66 (−71.45%). This ratio was shown to be the best predictor of cardiovascular disease in diabetic males in the Health Professionals' Follow-up Study (Jiang et al., 2004).

3.4. Summary of results

To recap, the purpose of this study was to evaluate the effects of *DG* on human anthropometric measurements, blood pressure levels, glucose, and lipid metabolism in obese and obese type 2 diabetic patients. The obtained results demonstrate the beneficial effects of *DG* on multiple markers of metabolic syndrome including weight, blood lipids, fasting blood glucose, etc. and thus may have a significant effect on cardiovascular health. We observed a significant reduction of weight: mean = −7.67 kg (−7.91%) and −6.04 kg (−5.97%) respectively in normoglycemic obese (OB) and type 2 diabetic (OB/DB) patients.

Table 1
The effect of *DG* on body weight, BMI, waist and hip circumference, and body fat in normoglycemic obese (OB) subjects.

Variables	Groups	T0	T4	T8	Change from baseline T8–T0 (%)
Weight (kg)	*DG*	98.43 ± 2.23	95.27 ± 2.26[a]	90.76 ± 2.32[b]	−7.67 ± 0.36 (−7.91)[c]
	Placebo	102.93 ± 2.8	102.13 ± 2.72	101.60 ± 2.81	−1.32 ± 0.21 (−1.31)
BMI (kg/m^2)	*DG*	38.09 ± 0.76	36.85 ± 0.77[a]	35.08 ± 0.77[b]	−3.00 ± 0.16 (−7.95)[c]
	Placebo	37.99 ± 1.23	37.7 ± 1.19	37.5 ± 1.22	−0.49 ± 0.08 (−1.30)
Waist (cm)	*DG*	107.87 ± 1.86	103.3 ± 1.9[a]	100.70 ± 1.95[b]	−7.17 ± 0.73[c] (−6.67)[c]
	Placebo	103.70 ± 2.61	102.43 ± 2.54	102.09 ± 2.55	−1.61 ± 0.21 (−1.54)
Hip (cm)	*DG*	127.87 ± 2.95	122.78 ± 2.42[a]	118.96 ± 2[b]	−8.91 ± 2.04 (−6.50)[c]
	Placebo	128.96 ± 2.52	127.7 ± 2.47	127.33 ± 2.42	−1.26 ± 0.2 (−1.32)
Body fat (%)	*DG*	45.64 ± 1.31	44.1 ± 1.35[a]	42.44 ± 1.26[b]	−3.20 ± 0.20 (−7.07)[c]
	Placebo	45.77 ± 1.16	45.67 ± 1.23	45.07 ± 1.19	−0.7 ± 0.11 (−1.57)

[a] $p<0.05$; compared with Placebo; adjusted for baseline.
[b] $p<0.001$; compared with Placebo; adjusted for baseline.
[c] $p<0.05$; compared with Initial; intragroup analysis.

Table 2
The effect of *DG* on body weight, BMI, waist and hip circumference, and body fat in obese type 2 diabetic (OB/DB) subjects.

Variables	Groups	T0	T4	T8	Change from baseline T8–T0 (%)
Weight (kg)	*DG*	99.84 ± 2.5	97.37 ± 2.29[a]	93.80 ± 2.22[b]	−6.04 ± 0.32 (−5.97)[c]
	Placebo	108.56 ± 2.47	108.24 ± 2.46	107.47 ± 2.54	−1.09 ± 0.24 (−1.06)
BMI (kg/m^2)	*DG*	38.21 ± 0.43	37.32 ± 0.37[a]	35.93 ± 0.35[b]	−2.28 ± 0.10 (−5.92)[c]
	Placebo	39.67 ± 1.08	39.58 ± 1.08	39.28 ± 1.09	−0.40 ± 0.09 (−1.03)
Waist (cm)	*DG*	109.78 ± 3.19	106.52 ± 3.00	103.70 ± 2.96[b]	−6.09 ± 0.60 (−5.49)[c]
	Placebo	109.22 ± 1.90	107.7 ± 1.80	107.39 ± 1.83	−1.83 ± 0.32 (−1.64)
Hip (cm)	*DG*	125.09 ± 1.59	122.52 ± 1.31[a]	121.96 ± 1.32[a]	−3.13 ± 0.39 (−2.45)[c]
	Placebo	132.52 ± 2.41	130.87 ± 2.34	130.52 ± 2.23	−2 ± 0.24 (−1.46)
Body fat (%)	*DG*	47.8 ± 0.69	46.64 ± 0.78[a]	45.53 ± 0.68[b]	−2.26 ± 0.20 (−4.74)[c]
	Placebo	47.69 ± 1.02	47.77 ± 1.13	47.13 ± 1.02	−0.55 ± 0.13 (−1.16)

[a] $p<0.05$; compared with Placebo ; adjusted for baseline.
[b] $p<0.001$; compared with Placebo; adjusted for baseline.
[c] $p<0.05$; compared with Initial; intragroup analysis.

Table 3
Effect of *DG* on systolic (SBP) and diastolic (DBP) blood pressure in normoglycemic obese (OB) subjects.

Variables	Groups	T0	T4	T8	Change from baseline T8–T0 (%)
SBP (mmHg)	*DG*	146.52 ± 1.33	134.52 ± 2.04[a]	133.43 ± 1.96[a]	−13.09 ± 1.95 (−8.86)[b]
	Placebo	138.09 ± 3.86	137.43 ± 3.38	134.87 ± 3.53	−3.22 ± 1.07 (−2.15)
DBP (mmHg)	*DG*	92.52 ± 2.19	82.22 ± 2.10[a]	80.7 ± 2.33[a]	−10.3 ± 1.45 (−12.74)[b]
	Placebo	86.3 ± 1.69	85.52 ± 1.69	84.96 ± 1.98	−0.78 ± 1.8 (−1.67)

[a] $p<0.001$; compared with Placebo; adjusted for baseline.
[b] $p<0.05$; compared with Initial; intragroup analysis.

Although significant for both groups, the reductions in BMI, waist and hip circumferences, and body fat were generally higher in normoglycemic obese participants (Tables 1 and 2).

4. Discussion

The results of the present study showed that *DG* is more effective in reducing the plasma glucose levels in both normoglycemic and type 2 diabetic obese patients compared to placebo. A previous study from our laboratory (Kuate et al., 2010) reported that the *in vitro* and *in vivo* antioxidant properties of *DG* extracts might be due to the synergistic effects of the multiple bioactive microcomponents (e.g., polyphenols and phenolic acids) present in this spice, which might maintain glucose levels by exerting their effects on glucose absorption, utilization, and excretion. These compounds are free radical scavengers and can improve the complications of diabetes such as glycation, glycoxidation, atherosclerosis and hyperlipidemia (Rukmini, 2000; Da Silva, Terezinha, Shirai, Terao, & Abdalla, 2008). The positive effects of polyphenols (and vitamin E) to correct the complications of diabetes due to their antioxidant property have been reported (Mock, 1996; Sharangi, 2009). Among antioxidants, α-lipoic acid, acting as lipoate, has also gained attention as a potential therapeutic agent for diabetes-induced complications (Suzuki, Tsuchiya & Packer, 1992).

Non-enzymatic glycation of proteins has been found to increase in a variety of proteins, such as hemoglobin, in diabetic patients. Lipoate prevents glycation and subsequent glucose-induced structural modification of various proteins (Suzuki et al., 1992). Cardiovascular complications, characterized by endothelial dysfunction and accelerated atherosclerosis, are the leading cause of morbidity and mortality associated with diabetes. There is growing evidence that excess generation of highly reactive free radicals, largely due to hyperglycemia, causes oxidative stress, which further exacerbates the development and progression of diabetes and its complications (Boden & Shulman, 2002). It is becoming increasingly clear that ameliorating oxidative stress through treatment with antioxidants like those present in DG might be an effective strategy for reducing diabetic complications. The increased glycosylated hemoglobin in the diabetic patients indicates that erythrocytes were more prone to oxidative stress in diabetes (Bunn, 1981). Glycosylated hemoglobin had been found to increase in patients with diabetes mellitus (Baskaran, Kizar, Radha, & Shanmugasundaram, 1990).

In this study, oral administration of *DG* decreased hyperglycemia, hence the level of HbA1c decreased similar to results observed in other studies (Koenig, Peterson, Jones, Saudek, Lehrman, & Cerami, 1976). As previously reported (Yesilbursa et al., 2005), the current study also showed that daily consumption of 800 mg of *DG* for 8 weeks led to a reduction of fasting blood glucose levels accompanying weight loss in obese subjects. A decrease in plasma glucose, coupled with an improvement in lipid profile, might also ensure protection against lipid plaque buildup and rupture in artery walls. Such protection

Table 4
Effect of *DG* on systolic (SBP) and diastolic (DBP) blood pressure in obese type 2 diabetic OB/DB) subjects.

Variables	Groups	T0	T4	T8	Change from baseline T8–T0 (%)
SBP (mmHg)	*DG*	137.22 ± 2.13	125.09 ± 1.02[a]	119.43 ± 1.37[b]	−17.78 ± 0.77 (−12.83)[c]
	Placebo	141.22 ± 4.31	142.09 ± 3.94	139.52 ± 3.17	−1.7 ± 1.97 (−0.50)
DBP (mmHg)	*DG*	91.7 ± 1.99	82.7 ± 1.52[a]	76.43 ± 2.33[b]	−15.26 ± 1.72 (−16.62)[c]
	Placebo	92.87 ± 1.56	93.3 ± 1.49	93.09 ± 1.59	0.22 ± 0.79 (+0.30)

[a] $p<0.05$; compared with Placebo; adjusted for baseline.
[b] $p<0.001$; compared with Placebo; adjusted for baseline.
[c] $p<0.05$; compared with Initial; intragroup analysis.

Table 5
The effect of *DG* on blood glucose, triglyceride(TAG), blood total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), and the TC/HDL-C ratio in obese (OB) subjects.

Variables	Groups	T0	T4	T8	Change from baseline T8–T0 (%)
Glucose (mg/dL)	*DG*	106.7 ± 2.06	85.09 ± 3.55[a]	77.78 ± 2.97[b]	−28.91 ± 7,21 (−26.01)[c]
	Placebo	99.87 ± 1.33	104.43 ± 1.82	99.78 ± 2.61	−0.09 ± 10.36 (−0.13)
TAG (mg/dL)	*DG*	123.65 ± 6.25	71.78 ± 9.13[a]	68.22 ± 4.58[b]	−55.43 ± 7.08 (−41.31)[c]
	Placebo	116.26 ± 9.92	116.57 ± 7.82	114.7 ± 6.86	−1.57 ± 7.74 (+10.64)
TC (mg/dL)	*DG*	174.87 ± 6.46	139.29 ± 7.29[a]	131.77 ± 5.79[b]	−43.09 ± 6.13 (−23.73)[c]
	Placebo	166.63 ± 6.62	169.41 ± 7.03	177.27 ± 10.27	10.63 ± 5.40 (+5.35)
HDL-C (mg/dL)	*DG*	36.22 ± 3.12	52.54 ± 3.24[a]	61.8 ± 2.76[b]	25.58 ± 4.93 (+126.06)[c]
	Placebo	37.36 ± 2.99	32.71 ± 2.97	31,87 ± 2.03	−5.49 ± 30 (+0.49)
LDL-C (mg/dL)	*DG*	111.07 ± 8.99	74.19 ± 7.7[a]	60.85 ± 6.4[b]	−50.22 ± 7.79 (−39.27)[c]
	Placebo	105.45 ± 7.72	111.33 ± 8.74	124.1 ± 9.13	18.66 ± 4.73 (+20.29)
TC/HDL-C	*DG*	5.89 ± 0.61	2.96 ± 0.29[a]	2.25 ± 0.18[b]	−3.64 ± 0.64 (−51.92)[c]
	Placebo	5.87 ± 0.91	6.62 ± 0.85	5.95 ± 0.4	0.08 ± 0.70 (−27.16)

[a] $p<0.05$; compared with Placebo; adjusted for baseline.
[b] $p<0.001$; compared with Placebo; adjusted for baseline.
[c] $p<0.05$; compared with Initial; intragroup analysis.

Table 6
The effect of *DG* on blood glucose, triglyceride(TAG), blood total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), and the TC/HDL-C ratio in obese type 2 diabetic (OB/DB) subjects.

Variables	Groups	T0	T4	T8	Change from baseline T8–T0 (%)
Glucose (mg/dL)	*DG*	193.3 ± 7.73	141.52 ± 6.48[a]	89.22 ± 2.74[a]	−104.09 ± 5.85 (−50.91)[b]
	Placebo	206.52 ± 9.39	201.70 ± 8.92	200.17 ± 8.27	−6.35 ± 3.05 (−2.47)
TAG (mg/dL)	*DG*	138.87 ± 3.13	47.17 ± 5.95[a]	59.78 ± 4.41[a]	−79.09 ± 5.92 (−56.10)[b]
	Placebo	150.74 ± 5.90	155.39 ± 6.69	152.3 ± 8.72	1.57 ± 6.87 (+1.03)
TC (mg/dL)	*DG*	240.49 ± 7.15	174.95 ± 3.16[a]	167.89 ± 1.98[a]	−72.6 ± 5.4 (−31.97)[b]
	Placebo	241.83 ± 9.49	245.47 ± 6.94	265.56 ± 5.21	23.72 ± 4.89 (+12.18)
HDL-C (mg/dL)	*DG*	27.33 ± 2.84	53.78 ± 3.56[a]	66.9 ± 2.73[a]	39.57 ± 2.9 (+155.88)[b]
	Placebo	27.34 ± 2.32	22.76 ± 1.55	30.23 ± 1.86	12.12 ± 2.53 (+24.42)
LDL-C (mg/dL)	*DG*	178.49 ± 6.34	111.74 ± 9.70[a]	93.72 ± 9.61[a]	−84.77 ± 5.62 (−49.56)[b]
	Placebo	182.6 ± 9.72	188.15 ± 7.85	204.35 ± 5.75	21.75 ± 5.40 (+17.22)
TC/HDL-C	*DG*	9.25 ± 0.45	3.22 ± 0.05[a]	2.59 ± 0.18[a]	−6.66 ± 0.37 (−71.45)[b]
	Placebo	10.05 ± 0.81	11.84 ± 0.79	9.68 ± 0.7	−0.37 ± 0.93 (+6.64)

[a] $p<0.001$; compared with Placebo; adjusted for baseline.
[b] $p<0.05$; compared with Initial; intragroup analysis.

reduces the risk for hypertension and cardiovascular disease, as suggested by the decrease in blood pressure in the *DG* patients. We cannot, however, exclude the presence of hypotensive molecules in the formulation, as evidenced by the strongly decreased values of systolic and diastolic blood pressure in both treated groups. Roth and Keller (1963) reported that the oral and parenteral administration of certain extracts of *DG* plant material to dogs had an immediate hypotensive action lasting several hours with no side effects.

Research on human nutrition has led to an awareness of the health benefits of dietary supplements. Over-the-counter remedies based on nutritional supplements are extremely popular, especially those addressing obesity and body composition. It is widely believed that supplements with a complex array of naturally occurring bioactive non-nutrients may confer significant long-term health benefits. Various hypotheses explaining the mechanism of the weight/fat loss, blood lipid and glucose lowering effect of *DG* include: (1) its high

Table 7
The effect of *DG* on glycosylated (HbA1c) hemoglobin in non diabetic obese (OB) subjects.

Variables	Groups	T0	T4	T8	Change from baseline T8–T0 (%)
HbA1c (%)	*DG*	5.24 ± 0.22	5.19 ± 0.21	5.14 ± 0.20	−0.10 ± 0.03 (−1.59)
	Placebo	5.27 ± 0.16	5.30 ± 0.15	5.26 ± 0.15	−0.01 ± 0.03 (−0.06)

Table 8
The effect of *DG* on glycosylated (HbA1c) hemoglobin in obese type 2 diabetic (OB/DB) subjects.

Variables	Groups	T0	T4	T8	Change from baseline T8–T0 (%)
HbA1c (%)	*DG*	10.49 ± 0.21	8.87 ± 0.21[a]	7.60 ± 0.14[b]	−2.89 ± 0.20 (−27.11)[c]
	Placebo	10.87 ± 0.29	10.81 ± 0.28	10.43 ± 0.24	−0.43 ± 0.08 (−3.78)

[a] $p<0.05$; compared with Placebo; adjusted for baseline.
[b] $p<0.001$; compared with Placebo; adjusted for baseline.
[c] $p<0.05$; compared with Initial; intragroup analysis.

polyphenol content, with reports that naturally-occurring polyphenols can inhibit pancreatic lipase, thereby influencing fat digestion and affecting energy intake (McDougall & Stewart, 2005; Birari & Bhutani, 2007; Moreno et al., 2003; Gondoin, Grussu, Stewart & McDougall, 2010); (2) The inhibitory effects of flavonoids and phenolic acids, such as o-coumaric acid and rutin on 3 T3-L1 adipocytes, as indicated by the decrease in intracellular triglyceride content and GPDH activity have been elucidated by Hsu and Yen (2007); (3) It could be mediated through the down-regulated expression of adipogenic transcription factors—peroxisome proliferator-activated receptor γ and CCAAT/enhancer-binding proteins and adipocyte-specific proteins (leptin)—and then the up-regulated expression of adiponectin similar to *Irvingia gabonensis* extracts (Oben, Ngondi, & Blum, 2008). Further studies, however, are needed to understand the exact mechanism of action of *DG*.

In conclusion, the data from the present study confirms the hypothesis that *DG* improves weight loss and lipid profile diabetes control, reduces blood pressure, and may favor a reduction in cardiovascular disease risk in obese patients and individuals with type 2 diabetes. The reduction of glycosylated hemoglobin by this formulation is very useful for the control of blood glucose levels in diabetic subjects.

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