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Effect of grape seed extract on blood pressure in subjects with the metabolic syndrome

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Abstract

This study was undertaken to determine whether grape seed extracts (GSE) that contain powerful vasodilator phenolic compounds lower blood pressure in subjects with the metabolic syndrome. The subjects were randomized into 3 groups—(*a*) placebo, (*b*) 150 mg GSE per day, and (*c*) 300 mg GSE per day—and treated for 4 weeks. Serum lipids and blood glucose were measured at the beginning of the study and at the end. Blood pressure was recorded using an ambulatory monitoring device at the start of the treatment period and at the end. Both the systolic and diastolic blood pressures were lowered after treatment with GSE as compared with placebo. There were no significant changes in serum lipids or blood glucose values. These findings suggest that GSE could be used as a nutraceutical in a lifestyle modification program for patients with the metabolic syndrome.

1. Introduction

There is evidence that a diet rich in vegetables and fruit has a beneficial effect on blood pressure. This effect has been attributed to the presence of phenolic compounds in the plant products. These compounds have also been shown to have vasodilator effects [1]. Of all the phenolic compounds, those derived from grape seeds appear to have received the most attention, possibly because of their involvement with the French paradox [2]. These extracts have also been shown to activate endothelial nitric oxide synthase (eNOS) [1,3], up-regulate eNOS in cultured endothelial cells [4], and cause an endothelium-dependent relaxation (EDR) of blood vessels [3].

We have shown recently that an extract of grape seed (Meganatural BP; Polyphenolics, Fresno, CA) that is rich in polyphenolic compounds causes an EDR that is mediated by activation of the Akt/PI3 kinase signaling pathway resulting in phosphorylation of eNOS [3]. Abolition of the EDR by removal of antioxidant activity from the extract by methylation of the OH groups supported the contention that activation of eNOS is mediated through a redox-sensitive mechanism [5].

These data suggested that this extract had the potential to lower blood pressure in human subjects. Recent studies in a spontaneously hypertensive rat had demonstrated that polyphenols present in red wine lower blood pressure and enhance nitric oxide bioactivity without up-regulating eNOS [6]. The definition of the *metabolic syndrome* was that adopted by the National Cholesterol Education Program Adult Treatment Panel III [7]. It was diagnosed when 3 of the following factors were present: abdominal obesity, elevated serum triglycerides, low serum high-density lipoprotein (HDL) concentration, hypertension, and elevated blood glucose [8]. All these factors are associated with impaired endothelial function resulting from the combined effects of a decrease in systemic antioxidant activity and an increase in reactive oxygen/nitrogen species [9,10]. It has been shown that polyphenolics derived from

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grapes increase the antioxidant levels and improve the endothelial function [6,11]. Therefore, the investigation reported here was undertaken to examine the effect of this extract on blood pressure in subjects with the metabolic syndrome as proof of this concept. The study was a placebo-controlled double-blind trial.

2. Materials and methods

The study was approved by the Institutional Review Board of the University of California, Davis. The study was conducted on a sample of 27 adults (age, 25-80 years) with the metabolic syndrome [8]. The exclusion criteria were as follows: smokers (abstinence for <1 year); clinical evidence of coronary artery, pulmonary, gastrointestinal, or renal disease; and consumption of prescription medications and vitamin preparations.

After initial screening, the subjects were randomized into 3 groups to receive a placebo, 300 mg/d of grape seed extract (GSE), or 150 mg of GSE (Meganatural BP). The phenol content of this extract is 94%. A detailed analysis of the extract is archived at a previous publication [3]. Before starting treatment, they were fitted with an ambulatory blood pressure measurement device that recorded the daytime blood pressure at intervals of 1 hour over a 12-hour period (Model SE-25S; Sein Electronics, Koyang, South Korea). This system has been evaluated using a protocol approved by the British Hypertension Society (www.tiba.medical.com).

The subjects were advised to maintain their usual level of activity and diet. The latter was monitored by examining a 4-day food diary that was completed at the start and at the end of the study. After 4 weeks, a final 12-hour daytime ambulatory blood pressure was recorded. Fasting blood samples were collected for the following measurements at the start of the study and at the end: hemoglobin, white cell count with differential, serum lipids, chemistry panel including a serum glucose, insulin, and oxidized low-density lipoprotein (Ox-LDL). Additional samples were obtained from 5 subjects in the placebo group and in the group that received 300 mg/d for measurement of plasma catechin concentrations. These samples were obtained immediately before and 90 minutes after ingestion of a capsule.

Table 1
Baseline parameters in the 27 subjects

Parameters	Placebo group	150-mg/d group	300-mg/d group	Significance
Age (y)	46 ± 3	45 ± 3	47 ± 4	NS
Male/female	3/6	4/5	4/5	NS
Waist circumference (in)	43 ± 1.8	44 ± 1.7	42 ± 1.8	NS
BMI (kg/m^2)	36 ± 2.4	36 ± 1.4	37 ± 2.1	NS
Glucose (mg/dL)	98 ± 5.9	101 ± 11	105 ± 10	NS
Insulin (μU/L)	25 ± 4	26 ± 5	30 ± 4	NS
Ox-LDL (U/L)	58 ± 4	60 ± 6	62 ± 7	NS

NS indicates not significant; BMI, body mass index.

Table 2
Initial and final blood pressures (in millimeters of mercury)

	Placebo		GSE (150 mg/d)		GSE (300 mg/d)	
	Systolic	Diastolic	Systolic	Diastolic	Systolic	Diastolic
Start	123 ± 4	74 ± 4	134 ± 5	83 ± 3	127 ± 4	78 ± 3
4 wk	121 ± 4	70 ± 4	123 ± 4	77 ± 2	116 ± 3	71 ± 3
n	9		9		9	
P[a]	.2	.1	.003	.01	.007	.007
Power	NS	NS	>0.8	0.8	[b]	>0.8

[a] Comparisons between initial and final blood pressures.
[b] Signed rank test.

The Ox-LDL concentration in plasma was measured using an mAb-4E6–based enzyme-linked immunosorbent assay (Mercodia, Uppsala, Sweden). Plasma insulin concentrations were measured using human-specific radioimmunoassay (Millipore, Billerica, MA). The plasma catechin concentrations were measured using a high-performance liquid chromatography technique [12] (“Acknowledgment”).

2.1. Statistical analysis

The primary end points were the mean daytime systolic and diastolic blood pressures. Secondary end points were the changes in serum lipids and Ox-LDL. Baseline values in the 3 groups were compared using a 1-way analysis of variance (ANOVA). The changes in blood pressure in the subjects in the 3 groups were also compared using a 1-way ANOVA. A 2-way ANOVA was used to compare the blood pressures at the start of the study and at the end to demonstrate a time effect.

3. Results

Twenty-seven subjects who met the criteria for the metabolic syndrome were randomized. The baseline clinical data are given in Table 1. There were no significant differences in baseline parameters in the subjects assigned to the 3 groups (ANOVA, $P > .05$).

At the end of 4 weeks, both systolic and diastolic blood pressures decreased in the groups that received GSE, whereas the group that received the placebo showed no effect (2-way ANOVA, $P < .05$). The changes in blood pressure in the 3 groups were also analyzed, and it was found that the effect of the placebo was significantly different from the effects of the 2 doses of GSE (ANOVA, $P < .05$). The effects of 150- and 300-mg/d doses were similar. The findings on blood pressure are summarized in Table 2 and Fig. 1. There were no significant changes in heart rate in the 3 groups.

There were no changes in the serum total, LDL, and HDL cholesterol values in 3 groups (Table 3). We observed decreased levels of Ox-LDL in the groups that consumed 150 and 300 mg/d of GSE, but these differences were not significant compared with the placebo group. However, the change in Ox-LDL appeared to be inversely related to the

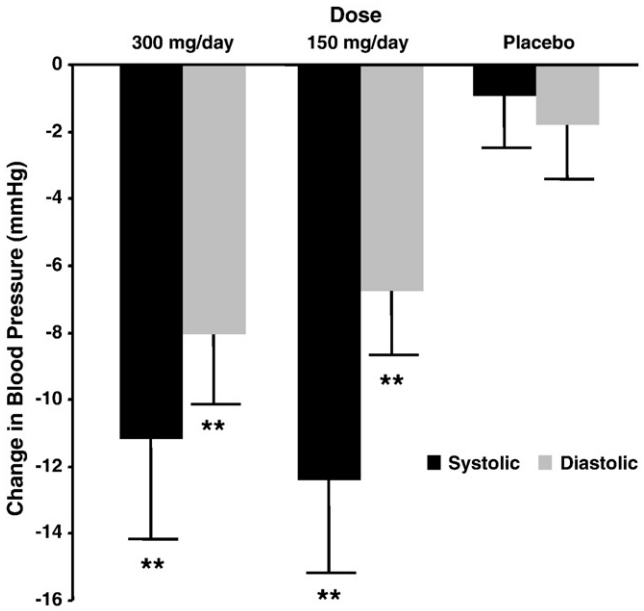

Fig. 1. Changes in blood pressure after administration of GSE for 4 weeks. Changes in systolic and diastolic blood pressure after 1 month of treatment. **Significantly different from corresponding placebo value ($P < .01$). Bars represent mean changes in the blood pressure ± SEM.

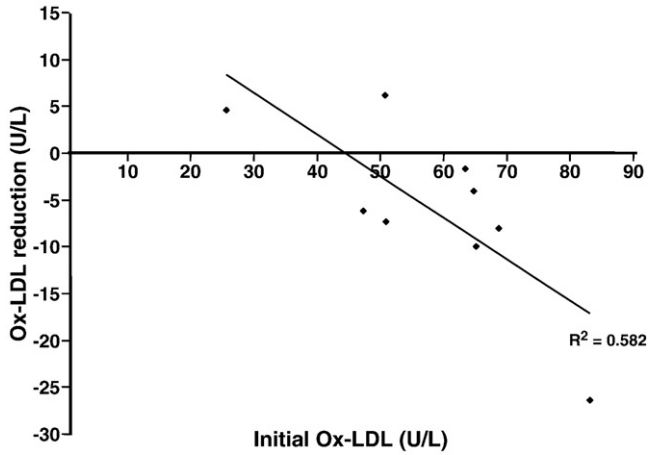

Fig. 2. Change in Ox-LDL levels plotted against the baseline Ox-LDL levels in plasma. Administration of GSE appeared to reduce the concentration of Ox-LDL in plasma in subjects with metabolic syndrome. This effect was particularly evident in those who received 300 mg/d of GSE. There was a significant positive correlation between the change in the concentration of Ox-LDL and the baseline Ox-LDL levels (n = 9, $P < .05$).

baseline (initial) concentration before starting treatment. This relationship was statistically significant in the group given 300 mg of GSE per day ($P < .05$) (Fig. 2).

The serum sodium, potassium, bicarbonate, creatinine, and blood urea nitrogen were also measured and were unchanged during the study. There were also no changes in the complete blood counts.

In 5 subjects who received the placebo capsules, there was no increase in total plasma catechin concentrations after 90 minutes (2.2 ± 3 vs 3.2 ± 3 ng/L, $P = .7$, paired t test). The corresponding values in the subjects who received 300 mg of the extract were 2.0 ± 4 and 22.0 ± 22.8 ng/L, respectively. This increase was significant ($P = .032$, Whitney-Mann).

4. Discussion

The findings of this randomized controlled trial indicate that GSE when administered at a dose of either 300 or 150 mg/d reduced both systolic and diastolic blood pressures in subjects with the metabolic syndrome as defined by the National Cholesterol Education Program Adult Treatment Panel III. All the blood pressures were obtained using ambulatory devices over 12-hour periods as opposed to a single clinic blood pressure measurement. These findings are consistent with several related observations.

We have demonstrated previously that the GSE used in the present study caused an EDR in the rabbit aorta [3]. It was shown also that, in human umbilical vein endothelial cells, eNOS was activated by GSE via the PI3 kinase/Akt pathway [3]. Wallerath et al [4] observed that GSE up-regulated eNOS in human umbilical vein endothelial cells. Based on these findings, it is suggested that the lowering of blood pressure in human subjects with metabolic syndrome after administration of the GSE is mediated by an eNOS-associated mechanism.

Grape seed extract used in the current study is known to have high content of polyphenolic compounds (94%). It is made up principally of catechin units and has an average polymerization of 2.4. The other unusual feature of this extract is the absence of terminal gallate units (see archived information in Edirisinghe et al [3]). The present study has

Table 3
Changes in serum lipids

	Placebo		150 mg/d		300 mg/d	
	Initial levels	After 1 mo	Initial levels	After 1 mo	Initial levels	After 1 mo
Total cholesterol (mg/dL)	199 ± 12	197 ± 11	216 ± 9	209 ± 10	194 ± 18	196 ± 15
HDL cholesterol (mg/dL)	51 ± 2	49 ± 3	51 ± 4	48 ± 4	44 ± 3	45 ± 3
LDL cholesterol (mg/dL)	125 ± 10	124 ± 9	147 ± 5	135 ± 10	126 ± 15	128 ± 14
Triglycerides (mg/dL)	179 ± 21	183 ± 19	176 ± 15	178 ± 15	178 ± 18	176 ± 15
Ox-LDL (U/L)	58 ± 4	60 ± 4	60 ± 6	57 ± 7	62 ± 7	58 ± 8

There is no significant difference before and after GSE treatments.

also shown that the extract is absorbed into the systemic circulation. Previous studies undertaken in humans have not shown a reduction in blood pressure. Some of these discrepancies may in part be due to differences in the composition of the extract [13].

The metabolic syndrome is an important public health problem affecting Western societies. In the United States, the prevalence of the condition is estimated to be 39% [14]. It is an important risk factor for the development of both coronary artery disease [15] and type 2 diabetes mellitus [16]. One of the associated features of the syndrome is the state of oxidative stress that is present in these individuals. One manifestation of this state is the increase in Ox-LDL in plasma that is a "precursor" to the development of atherosclerosis [17,18]. We observed that GSE at a dose of 300 mg/d reduced Ox-LDL, particularly when the Ox-LDL concentrations were high. It is suggested that this observation may point toward a potential therapeutic benefit from the extract. The observation in this article relating to blood pressure in humans is consistent with recent observations in spontaneously hypertensive rats treated with red wine polyphenols [6]. However, it was suggested that the mechanism involved was an enhancement in the bioactivity of nitric oxide through a reduction in oxidative stress.

In summary, the present study has demonstrated that an extract of grape seed lowers blood pressure in subjects with the metabolic syndrome. We have shown that the phenolic compounds in the extract are absorbed and that its antioxidant properties could reduce the concentration of Ox-LDL in plasma.

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References

[1] Cishek MB, Galloway MT, Karim M, et al. Effect of red wine on endothelium-dependent relaxation in rabbits. Clin Sci (Lond) 1997;97:507-11.

[2] De Lorgeril M, Salen P, Paillard F, et al. Mediterranean diet and the French paradox: two distinct biogeographic concepts for one consolidated scientific theory on the role of nutrition in coronary artery disease. Cardiovasc Res 2002;54:503-15.

[3] Edirisinghe I, Burton-Freeman B, Kappagoda T. The mechanism of the endothelium dependent relaxation evoked by a grape seed extract. Clin Sci (Lond) 2008;114:331-7.

[4] Wallerath T, Li H, Gödtel-Ambrust U, et al. Resveratrol, a polyphenolic phytoalexin present in red wine, enhances expression and activity of endothelial nitric oxide synthase. Circulation 2002;106:1652-8.

[5] Ndiaye M, Chataigneau M, Lobysheva I, et al. Red wine polyphenol-induced, endothelium-dependent NO-mediated relaxation is due to the redox-sensitive PI3-kinase/Akt-dependent phosphorylation of endothelial NO-synthase in the isolated porcine coronary artery. FASEB J 2004;19:455-7.

[6] López-Sepúlveda R, Jiménez R, Romero M, et al. Wine polyphenols improve endothelial function in large vessels of female spontaneously hypertensive rats. Hypertension 2008;51:1088-95.

[7] Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults. Executive Summary of The Third Report of The National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, And Treatment of High Blood Cholesterol In Adults (Adult Treatment Panel III). JAMA 2001;285:2486-97.

[8] Chobanian AV, Bakris GL, Black HR, et al. Seventh report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure. Hypertension 2003;42:1206-52.

[9] Suzuki T, Hirata K, Elkind MS, et al. Metabolic syndrome, endothelial dysfunction, and risk of cardiovascular events: the Northern Manhattan Study (NOMAS). Am Heart J 2008;156:405-10.

[10] Anter E, Keaney Jr JF. Oxidative stress, antioxidants, and endothelial function. Curr Med Chem 2004;11:1093-104.

[11] Leifert WR, Abeywardena MY. Cardioprotective actions of grape polyphenols. Nutr Res 2008;28:729-37.

[12] Gu L, House SE, Rooney LW, et al. Sorghum extrusion increases bioavailability of catechins in weanling pigs. J Agric Food Chem 2008;56:1283-8.

[13] Ward NC, Hodgson JM, Croft KD, et al. The combination of vitamin C and grape-seed polyphenols increases blood pressure: a randomized, double-blind, placebo-controlled trial. J Hypertens 2005;23:427-34.

[14] Grundy SM, Brewer Jr HB, Cleeman JI, et al. American Heart Association, National Heart, Lung, and Blood Institute. Definition of metabolic syndrome: report of the National Heart, Lung, and Blood Institute/American Heart Association conference on scientific issues related to definition. Circulation 2004;109:433-8.

[15] Malik S, Wong ND, Franklin SS, et al. Impact of the metabolic syndrome on mortality from coronary heart disease, cardiovascular disease, and all causes in United States adults. Circulation 2004;110:1245-50.

[16] Laaksonen DE, Lakka HM, Niskanen LK, et al. Metabolic syndrome and development of diabetes mellitus: application and validation of recently suggested definitions of the metabolic syndrome in a prospective cohort study. Am J Epidemiol 2002;156:1070-7.

[17] Sigurdardottir V, Fagerberg B, Hulthe J. Circulating oxidized low-density lipoprotein (LDL) is associated with risk factors of the metabolic syndrome and LDL size in clinically healthy 58 year old men (AIR study). J Internal Med 2002;252:440-7.

[18] Holvoet P, Vanhaecke J, Janssens S, et al. Oxidized LDL and malondialdehyde-modified LDL in patients with acute coronary syndromes and stable coronary artery disease. Circulation 1998;98:1487-94.