

Reduction of cardiovascular risk factors in subjects with type 2 diabetes by Pycnogenol supplementation

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Abstract

Patients with type 2 diabetes are at considerable risk of excessive morbidity and mortality from cardiovascular disease (CVD). We investigated the clinical effectiveness of Pycnogenol, a flavonoid-rich dietary supplement, in reducing antihypertensive medication use and CVD risk factors in subjects with type 2 diabetes. Forty-eight individuals were enrolled in a randomized, double-blind, placebo-controlled trial with parallel-group design. Patients were diagnosed with both type 2 diabetes and mild to moderate hypertension and were undergoing treatment with angiotensin-converting enzyme (ACE) inhibitors. Subjects were randomly assigned to receive either Pycnogenol pill (125 mg daily) or matched placebo for 12 weeks. According to the values of blood pressure (BP) measured at 2-week intervals, the pretrial ACE inhibitor dosage was left unchanged, reduced by 50%, or brought back to the pretrial dosage until a stable BP was obtained. Fasting plasma glucose, low-density lipoprotein (LDL) cholesterol, glycosylated hemoglobin (HbA1c), serum endothelin-1, and urinary albumin were evaluated monthly. Pycnogenol treatment achieved BP control in 58.3% of subjects at the end of the 12 weeks with 50% reduction in individual pretrial dose of ACE-inhibitors ($P < .05$). Plasma endothelin-1 decreased by 3.9 pg/mL in Pycnogenol-treated group vs 0.5 pg/mL increase in control group ($P < .001$). Mean HbA1c dropped by 0.8% in Pycnogenol-treated group ($P < .05$), whereas it decreased by 0.1% in control group. Fasting plasma glucose declined by 23.7 mg/dL in Pycnogenol-treated group vs 5.7 mg/dL in control group ($P < .0001$). Low-density lipoprotein cholesterol improved significantly in Pycnogenol-treated group, declining by 12.7 mg/dL ($P < .001$). A significant decrease in urinary albumin level was observed at week 8 compared with the control group ($P < .05$). However, this reduction was not significant at 12th week. After 12 weeks of supplementation, Pycnogenol resulted in improved diabetes control, lowered CVD risk factors, and reduced antihypertensive medicine use vs controls.

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Abbreviations: CVD, cardiovascular disease; ACE inhibitors, angiotensin-converting enzyme inhibitors; HbA1c, glycosylated hemoglobin; LDL-C, low-density lipoprotein-cholesterol; BP, blood pressure; NO, nitric oxide.

1. Introduction

Diabetes mellitus affects approximately 20 million people in the United States, approximately 7% of the US population [1]. It is expected to increase by 50% to 70%

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Table 1
Baseline demographic and characteristics of the study intention to treat groups

Variable	Placebo	Pycnogenol
Number	24	24
Age (y)	58.4 ± 11.5	61.3 ± 9.1
Gender (male:female)	14:10	13:11
Duration of diabetes (y)	14.2 ± 8.5	12.9 ± 11.6
Pretrial systolic BP (mm Hg)	137.0 ± 1.0	139.0 ± 1.3
Pretrial oral antidiabetic medications (%)		
First-generation sulfonylureas	0	6
Second-generation sulfonylureas	38	25
Metformin	38	44
Thiazolidinediones	19	25

Data are mean ± SEM or percentage. Analyzed by Student *t* test.

within the next 25 years. Numerous epidemiologic studies have demonstrated that type 2 diabetes increases the risk of cardiovascular morbidity and mortality [2]. The increased risk is due to the detrimental vascular effects of prolonged exposure to a hyperglycemic environment as well as the higher prevalence of associated cardiovascular risk factors: atherosclerosis, hypertension, and clotting abnormalities. Diabetes mellitus substantially increases the risk of developing atherosclerosis [3]. Hypertension is a common comorbid condition of diabetes, affecting most patients with diabetes. Elevated blood pressure (BP) contributes to diabetic microvascular and macrovascular complications [4]. This concomitant presence of hyperglycemia, dyslipidemia, and hypertension magnifies the risk of cardiovascular disease and requires aggressive management of all cardiovascular risk factors. Treatment of hypertension and dyslipidemia in diabetic patients produces substantial decreases in cardiovascular and microvascular diseases [5]. However, hypertension associated with diabetes has certain pathophysiologic characteristics that provide clinical challenges. It is difficult to achieve the target BP goal, which is lower in diabetics, with pharmacologic therapy without incurring undesirable side effects. In addition, such drugs are expensive and only somewhat effective in treating other cardiovascular risk factors, thus encouraging investigation of antihypertensive dietary supplements as therapy.

Pycnogenol, a standardized extract from the bark of French maritime pine (*Pinus maritima*) with significant antioxidant activity, primarily comprises phenolic compounds (catechin, epicatechin, and taxifolin) and flavonoids (procyanidins) [6]. It has been documented that Pycnogenol mediates a number of beneficial effects on the cardiovascular system. Previous studies have shown that Pycnogenol supplementation is associated with reducing platelet aggregation [7], lowering low-density lipoprotein cholesterol (LDL-C) and increasing high-density lipoprotein cholesterol [8], and modifying hypertension [9]. Furthermore, Pycnogenol, in a dose-dependant manner, has been shown to reduce fasting and postprandial serum glucose levels [10] and to lower glycosylated hemoglobin

(HbA1C) [11] in patients with type 2 diabetes mellitus. Therefore, we hypothesized that the Pycnogenol, a novel mixture of flavonoids, may reduce the antihypertensive medicine use and cardiovascular risk factors in individuals with type 2 diabetes.

2. Methods and materials

2.1. Human subjects

The study population consisted of men and women, 40 to 75 years of age, with noninsulin-dependent type 2 diabetes. They were receiving pharmaceutical treatment (angiotensin-converting enzyme [ACE] inhibitors) for hypertension with pretrial systolic BP of 130 to 150 mm Hg. Exclusion criteria included the following: having type 1 diabetes; using insulin; taking any supplements other than single daily multivitamin; having any major illness such as cancer, asthma, or heart failure; having any previous cardiac events; being pregnant; or being nursing mother.

2.2. Study design

The protocol of this 12-week, randomized, double-blind, placebo-controlled trial was approved by the institutional review board at the University of Arizona and performed in accordance with the Declaration of Helsinki. Study subjects were recruited from newspaper advertisements. At the screening visit, all subjects gave written informed consent before participating in this research trial. At the second visit, subjects were randomly assigned to receive either Pycnogenol pill (25 mg, 5 times a day) or matched placebo, which remained constant

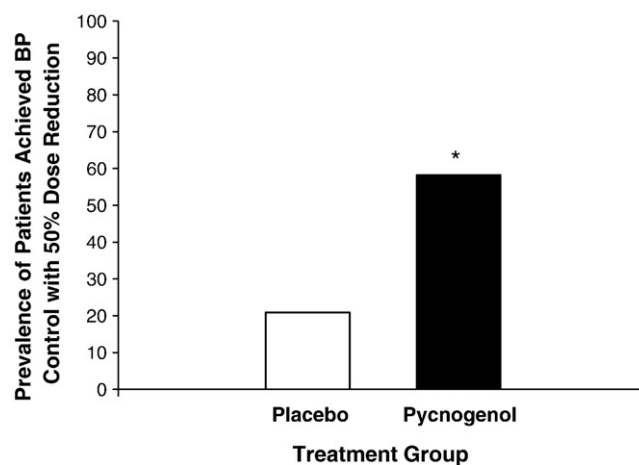


Fig. 1. Percentage of study subjects who could achieve BP control with 50% reduction in individual pretrial dose of ACE inhibitors, after 12-week supplementation with Pycnogenol. Blood pressure was evaluated at baseline and at biweekly follow-up visits. The pretrial ACE inhibitor dosage was either left unchanged, reduced by 50%, or brought back to the pretrial dosage until a stable BP was obtained. Values are means (n = 24 subjects in each group). Analyzed by χ^2 test with Yates correction, **P* < .05 compared with the placebo group.

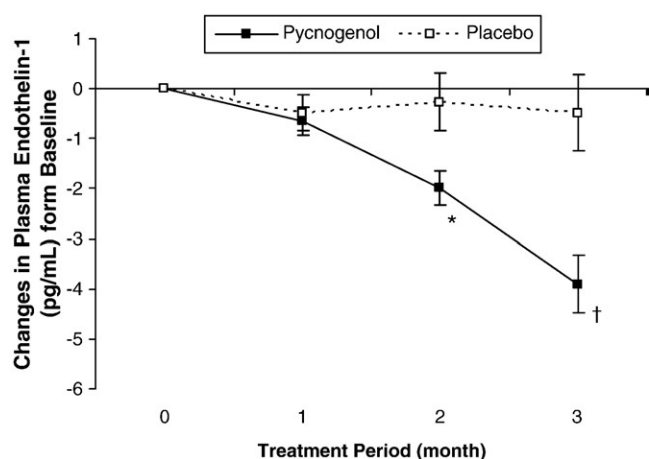


Fig. 2. Changes in plasma endothelin-1 level from baseline overtime, after 12-week supplementation with Pycnogenol in subjects with type 2 diabetes. Values are means \pm SEM ($n = 24$ subjects in each group). Analyzed by Student t test, * $P < .05$ and † $P < .001$ compared with the placebo group.

throughout the study period. Study participants continued the ACE inhibitors and other current medications, which included a variety of antidiabetic medications (Table 1).

Blood pressure and heart rate were recorded at baseline and at biweekly follow-up visits thereafter for 12 weeks. Blood pressure and heart rate were measured on the left arm after 10-minute rest. Korotkoff sounds I and V were taken as the systolic and diastolic BPs, respectively. Repeated readings were taken at 2-minute intervals, for a total of 3 sitting measurements, and the average was recorded. According to the values of systolic BP measured at 2-week intervals, the pretrial ACE inhibitor dosage was either left unchanged, reduced by 50%, or brought back to the pretrial dosage until a stable BP was obtained. All other treatments, including antidiabetic medications, remained constant throughout the study period. At the monthly follow-up visits, unused pills were collected and counted to assess compliance. Changes in concomitant medications and clinical adverse events at follow-up visits were searched for by questioning, and none was reported.

Table 2

The effect of Pycnogenol on plasma endothelin-1, glucose, HbA1c, serum LDL-C, and urinary albumin over time in subjects with type 2 diabetes

Variable	Treatment period							
	Baseline		Week 4		Week 8		Week 12	
	Placebo	Pycnogenol	Placebo	Pycnogenol	Placebo	Pycnogenol	Placebo	Pycnogenol
Endothelin-1 (pg/dL)	23.0 \pm 1.6	21.9 \pm 1.5	22.5 \pm 2.1	21.3 \pm 2.4	22.7 \pm 1.8	19.9 \pm 1.8	22.5 \pm 1.5	18.0 \pm 1.7 [†]
Glucose (mg/dL)	151.1 \pm 12.1	142.3 \pm 9.8	149.7 \pm 10.6	134.3 \pm 9.2	143.4 \pm 13.0	123.2 \pm 8.6	145.3 \pm 11.4	118.6 \pm 9.5
HbA1c (%)	8.1 \pm 0.4	7.9 \pm 0.3	8.2 \pm 0.3	7.8 \pm 0.3	8.1 \pm 0.3	7.3 \pm 0.2	8.0 \pm 0.4	7.1 \pm 0.2 ^{*†}
LDL-C (mg/dL)	104.0 \pm 5.1	106.4 \pm 6.3	103.4 \pm 5.7	105.3 \pm 5.6	101.1 \pm 7.1	94.8 \pm 9.1	107.0 \pm 4.8	93.7 \pm 6.2
Urinary albumin (mg/L)	37.4 \pm 7.0	29.6 \pm 5.9	40.0 \pm 6.5	30.9 \pm 5.1	42.6 \pm 7.1	20.4 \pm 3.7 [†]	36.1 \pm 6.3	22.2 \pm 6.1

Measurements were performed on serum, plasma, and urine samples after 8 hours of fasting at the baseline and at monthly basis. Values are means \pm SEM ($n = 24$ subjects in each group). Analyzed by Student t test.

* $P < .05$ compared with the baseline.

† $P < .05$ compared with the placebo group.

2.3. Serum measures

Change from baseline at weeks 4, 8, and 12 were assessed after 8 hours of fasting for plasma endothelin-1, serum LDL-C, HbA1c, and plasma glucose. Urinary albumin concentration was measured from spot urine samples using semiquantitative screening dipstick test on a monthly basis. All tests were 2 to 4 hours after the last consumption of the pills. Endothelin-1 level was quantified in plasma in duplicate samples by enzyme-linked immunoabsorbent assay (R&D Systems, Minneapolis, Minn). The VA hospital and J2 Laboratories (Tucson, Ariz) conducted standardized analyses for all other assays. Serum LDL-C was measured directly by the cholesterol esterase–cholesterol peroxidase coupling method, HbA1c by inhibition of latex agglutination, and plasma glucose by a double-enzyme assay with hexokinase and glucose-6-phosphate dehydrogenase, using an Olympus AU640 analyzer (Olympus America, Inc, Melville, NY).

2.4. Statistical methods

Analysis was performed according to the intention to treat principle. Thus, all randomized patients who received at least 1 dose of study treatment and who had both a baseline and at least 1 postbaseline measurement were analyzed. The data are expressed as mean \pm SEM. Statistical analyses were performed with SPSS version 11.5 (SPSS Institute, Chicago, Ill) [12]. Chi-square test with Yates correction was used for noncontinuous variables for the prevalence study. Student t test was used to assess the statistical significance of the continuous variables. Comparable nonparametric test (Mann-Whitney U test) was substituted when tests for normality and equal variance failed [13]. A value of $P < .05$ was used as a criterion for statistical significance.

3. Results

Of 48 subjects who met the inclusion and exclusion criteria, 1 subject was withdrawn due to unwanted side effect (placebo group) and 2 were lost to follow-up (1 in Pycnogenol and 1 in placebo group). As shown in Table 1,

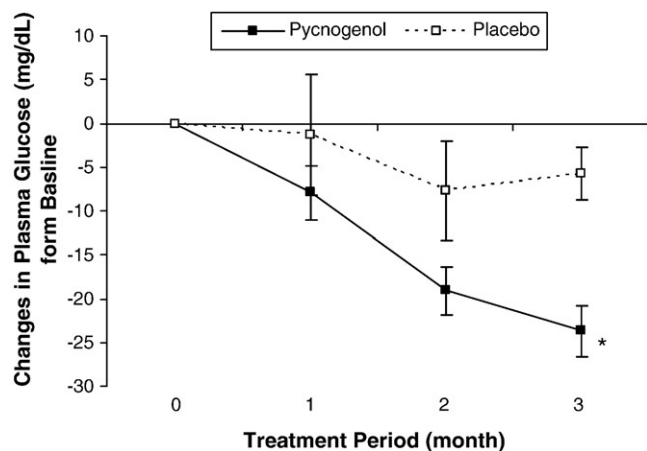


Fig. 3. Changes in fasting plasma glucose level from baseline overtime, after 12-week supplementation with Pycnogenol in subjects with type 2 diabetes. Values are means \pm SEM ($n = 24$ subjects in each group). Analyzed by Student t test, * $P < .0001$ compared with the placebo group.

demographic and clinical characteristics did not differ significantly between the 2 groups. The initial systolic BP was comparable (137.0 ± 1.0 vs 139.0 ± 1.3 mm Hg). Thus, 58.3% of the Pycnogenol-treated subjects achieved BP control at the end of the 12 weeks, with a 50% reduction in individual pretrial dose of ACE inhibitors ($P < .05$). However, in the placebo group, only 20.8% of subjects maintained control (Fig. 1). Concomitantly, a marked reduction of 3.9 pg/mL in plasma endothelin-1 level occurred in Pycnogenol group compared with control group ($P < .001$; Fig. 2).

Pycnogenol supplementation resulted in the improvement of all serum variables and urinary albumin, which were significant at 8th week of treatment for urinary albumin and at 12th week for endothelin-1 and HbA1c, as compared with the placebo group ($P < .05$; Table 2). Subjects treated with

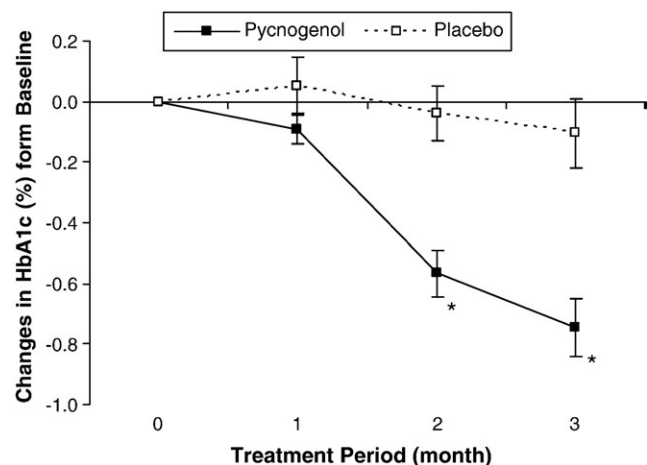


Fig. 4. Changes in HbA1c level from baseline overtime, after 12-week supplementation with Pycnogenol in subjects with type 2 diabetes. Values are means \pm SEM ($n = 24$ subjects in each group). Analyzed by Student t test, * $P < .001$ compared with the placebo group.

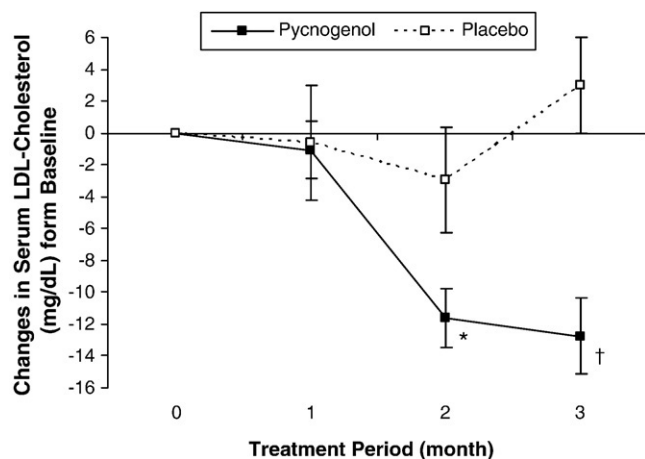


Fig. 5. Changes in serum LDL-C level from baseline overtime, after 12-week supplementation with Pycnogenol in subjects with type 2 diabetes. Values are means \pm SEM ($n = 24$ subjects in each group). Analyzed by Student t test, * $P < .05$ and † $P < .001$ compared with the placebo group.

Pycnogenol demonstrated a reduction of 19.1 ± 2.7 and 23.7 ± 2.9 mg/dL in fasting plasma glucose at weeks 8 and 12, respectively. This change was only significant at week 12 when compared with the control group ($P < .0001$; Fig. 3). Moreover, Pycnogenol treatment was associated with a sustained decrease in HbA1c level, which was significantly lower at 8th and 12th weeks of the study, compared with the control group ($P < .001$; Fig. 4). At week 12, the mean HbA1c dropped from 7.9% to 7.1% in Pycnogenol-treated group, whereas the control group decreased nonsignificantly from 8.1% to 8.0%. A marked decrease in serum LDL-C of 11.6 ± 1.8 and 12.7 ± 2.3 mg/dL was observed in the group supplemented with Pycnogenol at the 8th and 12th weeks, respectively ($P < .05$ and $P < .001$, respectively; Fig. 5) when compared with the placebo group. Urinary albumin declined significantly more in the Pycnogenol-treated group

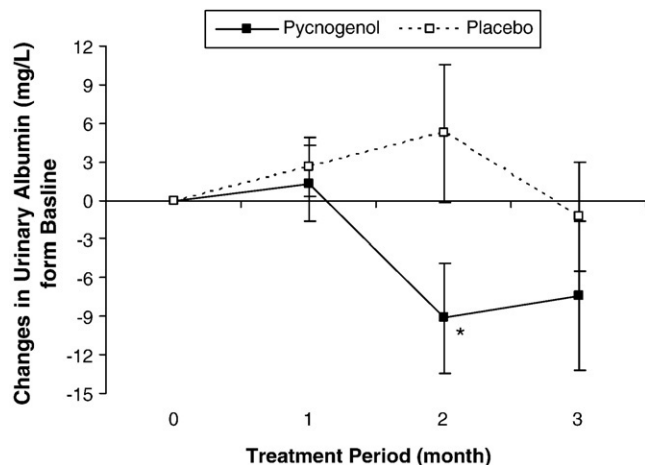


Fig. 6. Changes in urinary albumin level from baseline overtime, after 12-week supplementation with Pycnogenol in subjects with type 2 diabetes. Values are means \pm SEM ($n = 24$ subjects in each group). Analyzed by Student t test, * $P < .05$ compared with the placebo group.

compared with the control, dropping by 9.13 ± 4.26 and 7.39 ± 5.75 mg/L at weeks 8 and 12, respectively. These changes were only significant at week 8 when compared with the placebo group ($P < .05$; Fig. 6). Compliance measured by pill usage was similar in both groups: 94% in the Pycnogenol arm vs 96% in placebo arm.

4. Discussion

Research on human nutrition has led to an awareness of the health benefits of dietary supplements. Specifically, it has been recognized that the dietary supplements with complex array of naturally occurring bioactive nonnutrients may confer significant long-term health benefits. Pycnogenol is becoming another dietary supplement of bioactive nonnutritive molecules with health-promoting activities, important in understanding the role of supplements and foods in human diets and nutrition. The present study shows that Pycnogenol supplementation helps to maintain control of BP with a lower dose of ACE inhibitors in individuals with type 2 diabetes. Concomitantly, a marked decrease of 17.8% occurred in serum endothelin-1, an endothelial-derived vasoconstrictor peptide that plays an important role in modulating vascular tone. This finding is consistent with another study [11] that showed that the control of hypertension occurred with a lower dose of the calcium antagonist nifedipine in those patients receiving Pycnogenol supplementation and was associated with a 20% decrease in serum endothelin-1. Notably, the urinary concentration of albumin was reduced in Pycnogenol-treated subjects despite the reduction in pretrial ACE inhibitors dosage, providing evidence on the renoprotective effect of Pycnogenol which could reduce diabetic nephropathy.

Our findings indicate that the antihypertensive effect of Pycnogenol is mediated, at least in part, by suppression of serum endothelin-1, which is generally higher than normal in patients with type 2 diabetes and hypertension [14]. However, this significant antihypertensive effect could be based on several mechanisms operating simultaneously. One could be the inhibitory effect of Pycnogenol on ACE [15], which should reduce serum angiotensin-II level and improves flow-mediated vasodilation. Moreover, Pycnogenol stimulates the production of nitric oxide (NO) in isolated aortic rings [16]. Nitric oxide synthesized by endothelial NO synthase is a potent endothelium-derived vasorelaxant substance. However, in addition to being the main determinant of basal vascular smooth muscle tone, NO opposes the actions of potent endothelium-derived contracting factors such as angiotensin-II, endothelin-1, and reactive oxygen species [12]. In addition, Pycnogenol is reported to induce a further shift in balance away from vasoconstrictor factors such as thromboxane B2 [9] toward vasodilator factors such as 6 keto-prostaglandin-F1a, a prostacyclin's metabolite [11].

Moreover, the present study revealed that Pycnogenol supplementation resulted in improved diabetes control. Plasma fasting glucose and HbA1c levels gradually

improved and reached statistical significance at the 8th week for HbA1c and 12th week for fasting glucose when compared with placebo. In agreement with our results, other publications found the antidiabetic effect of Pycnogenol as evidenced by marked reduction of HbA1c and fasting glucose levels [10,17]. The glucose-lowering effect of Pycnogenol is dose-dependent and does not seem to be related to the enhancement of insulin secretion [10]. It might be mediated through the inhibition of α -glucosidase [18], thus diminishing glucose intestinal resorption.

Furthermore, this study has shown that Pycnogenol supplementation resulted in significant improvement in serum LDL-C level, another cardiovascular risk factor commonly seen in individuals with diabetes. Corresponding to our observation, several clinical trials have found lower LDL-C values after treatment with Pycnogenol [19,20]. Moreover, Pycnogenol has been found to protect LDL from oxidation [21] and to inhibit the inducible expression of ICAM-1 in human endothelial cells [22]. These changes with the hypolipidemic effect of Pycnogenol may reduce the risk of atherogenesis. The antioxidant activity of Pycnogenol [6] as well as its inhibitory effect on NF- κ B activation [23] and proinflammatory cytokine production [24] may also contribute to the protection afforded by Pycnogenol in the present study.

There are a few potential limitations to this study. The first limitation of our study concerns the very restricted inclusion and exclusion criteria (because of the target population being studied) that may limit the generalizability of the findings. Another limitation of the study was the relatively small sample size. Due to the limited size of the study population, we combined patients from different ethnic backgrounds in one group to achieve the required statistical power. Another limitation was that we did not differentiate between recently diagnosed diabetic patients and those who were diagnosed many years ago who may have been better controlled.

In summary, these data confirm the hypothesis that Pycnogenol improves diabetes control, reduces antihypertensive medicine use, and may favor a reduction in cardiovascular disease risk in individuals with type 2 diabetes.

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