



Pycnogenol[®], French maritime pine bark extract, improves endothelial function of hypertensive patients

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Abstract

A placebo-controlled, double-blind, parallel group study was performed with 58 patients to investigate effects of French maritime pine bark extract, Pycnogenol[®], on patients with hypertension. Supplementation of the patients with 100 mg Pycnogenol[®] over a period of 12 weeks helped to reduce the dose of the calcium antagonist nifedipine in a statistically significant manner. The intake of Pycnogenol[®] decreased endothelin-1 concentrations significantly compared to placebo while concentrations of 6-keto prostaglandin F_{1a} in plasma were significantly higher compared to placebo. Values for nitric oxide (NO) in plasma increased in both groups, but the differences were not significant. Angiotensin II concentrations in plasma were lowered in the placebo group to a larger extent than in the Pycnogenol[®] group. Heart rate, electrolytes and blood urea nitrogen were not changed during treatment in both groups of patients. Unwanted effects observed in both groups were of mild and transient nature, such as gastrointestinal problems, vertigo, headache and nausea. Differences in rate of side effects were not statistically significant between the two groups. Study results support a supplementation with Pycnogenol[®] for mildly hypertensive patients.

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Keywords: Pycnogenol[®] French maritime pine bark extract; Hypertension; Endothelin-1; Prostaglandin; Nifedipine

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Table 1
Patients data

Patients		N	Median	Percentile	
				25%	75%
age (years)	Placebo	30	58	46	66
	Pycnogenol®	28	56	46	64
stature (cm)	Placebo	30	164	160	171
	Pycnogenol®	28	168	162	172
weight (kg)	Placebo	30	68	62	75
	Pycnogenol®	28	69	64	76
sex	Placebo	30	male 18 female 12		
	Pycnogenol®	28	male 15 female 13		
pre-treatment-medication	Placebo	ACEI 10		CTM 5	
	Pycnogenol®	ACEI 13		CTM 4	
	Placebo	CCB + ACEI 6		CCB 7	
	Pycnogenol®	CCB + ACEI 4		CCB 5	

ACEI Angiotensin Converting Enzyme Inhibitor, CTM Chinese Traditional Medicine, CCB Calcium Channel Blocker.

Introduction

Pycnogenol®¹, a standardized extract from the bark of the French maritime pine (*Pinus pinaster Ait.*), consists of a concentrate of polyphenols. Main constituents are procyanidins, pharmacologically active biopolymers composed from units of catechin and epicatechin. Additionally, Pycnogenol® contains the bioflavonoids catechin and taxifolin and a number of phenolic acids (Rohdewald, 2002).

In vitro experiments demonstrated that Pycnogenol® and especially the procyanidins contained in it inhibit the angiotensin converting enzyme (ACE) (Blazso et al., 1996). In experiments with hypertensive rats Pycnogenol® dose-dependently decreased systolic and diastolic blood pressure after i.v. injection (Blazso et al., 1996). In a double blind, placebo controlled, cross-over study with hypertensive patients, oral administration of Pycnogenol® reduced systolic blood pressure from mildly hypertensive (140–159 mmHg) to normal values, but the reduction of diastolic blood pressure did not reach statistical significance (Hosseini et al., 2001). In the same study, a statistically significant lowering of the vasoconstrictor thromboxane B2 level was noted. However, hypertension could conceivably be reduced by Pycnogenol® by other mechanisms in addition to inhibition of ACE or thromboxane B2. The pine bark extract was able to stimulate in vitro the production of nitric oxide, thus counteracting the vasoconstriction by adrenaline or noradrenaline in isolated aortic rings from rats (Fitzpatrick et al., 1998). Because NO regulates vascular tonus, the enhancement of NO production could contribute to a lowering of blood pressure. Aim of the present study was to test whether a supplementation with Pycnogenol® could help to reduce the dose of an antihypertensive drug in patients with hypertension in a placebo-controlled double blind design.

¹ Pycnogenol®, distributed by Horphag Research Ltd.

Table 2

Dose of nifedipine after 12 weeks of dose adjustment

Dose		10 mg	15 mg	20 + 30 mg	p (Yates)
Placebo	number of patients	4	17	9	0,001
Pycnogenol®		16	6	6	

Chi-square test with Yates correction.

Patients and methods

Subjects

Fifty eight hypertensive patients (33 male, 25 female) with an average age of 57 years participated in the study. Patients were randomized to receive either 100 mg Pycnogenol® or placebo. The two groups of patients were not statistically different with respect to age, height, weight, sex and pretreatment medication against hypertension (Table 1).

Exclusion criteria included pregnancy, or nursing mothers, and malignant forms of hypertension. Use of vitamins was not allowed during the period of the study. Diagnosis and classification of hypertension was based on the hypertension inventory of the WHO (WHO/ISH, 1999). All patients signed informed consent forms. Patients were out-patients from three hospitals: Guang An men Hospital of Chinese Medical Science Research Institute in Beijing, Huang Si Hospital of the General Logistics Department of the Chinese People's Liberation Army in Beijing, Lai Wu Hospital of Xin Wen Mineral Bureau in ShanDong.

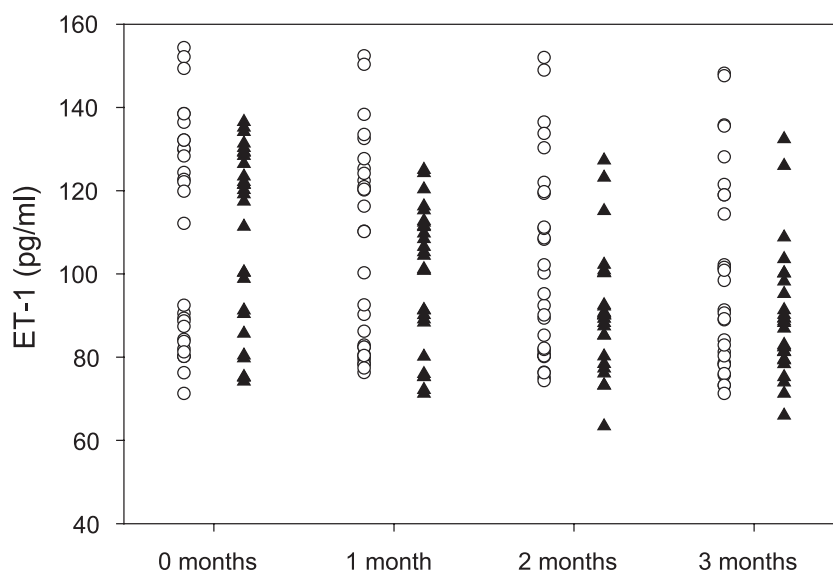


Fig. 1. Endothelin-1 values in plasma 0, 1, 2 and 3 months after start of treatment with placebo (white circles) and Pycnogenol® (black triangles).

Table 3

The percentage of endothelin-1 decrease in plasma after treatment with placebo and Pycnogenol® relative to pre-treatment values

Duration of treatment	Medication	N	Percent (median)	Percentile		p _{Bon}
				25%	75%	
1 month	Placebo	30	-1.91	-4.98	-0.80	<0.01
	Pycnogenol®	28	-11.04	-16.99	3.78	
2 months	Placebo	30	-3.14	-9.20	-1.94	<0.05
	Pycnogenol®	28	-20.37	-30.19	-6.21	
3 months	Placebo	30	-4.53	11.17	-2.76	ns
	Pycnogenol®	28	-21.08	-32.67	-6.74	

Mann-Whitney Rank Sum Test with Bonferoni correction: ns: not significant, *: p_{Bon} < 0.05, **: p_{Bon} < 0.01.

Methods

A randomized, double-blind, placebo-controlled, parallel group study was conducted. All patients received placebo for 2 weeks instead of their previous medication to confirm diagnosis. Thereafter one group of patients was treated with a calcium antagonist (nifedipine, sustained release tablets 5 mg, Shanghai Pharmaceuticals Co, Ltd.) plus placebo, while the other group was treated with calcium antagonist plus Pycnogenol®. All patients received 20 mg nifedipine at the start of study period. The dose of nifedipine was adjusted at 2 week intervals. According to the values of blood pressure measured at 2 week intervals, doses were either reduced or increased by 5 mg nifedipine until a stable blood pressure was obtained. Total treatment period was 12 weeks.

Blood pressure and heart rate were recorded at the start of the study and at 2 week intervals thereafter.

The following values were recorded in monthly intervals: angiotensin II, endothelin-1, 6-keto-prostaglandin F1 and nitrogen monoxide. Electrolytes, creatinine and blood urea nitrogen were monitored at the start and at the end of the study.

Blood pressure was measured at 8.30 a.m., 90 min after intake of medication. Patients rested for 15 min, followed by measurement in the sitting upright position. The mean of two measurements was recorded.

Table 4

The percentage of increase of 6-keto-prostaglandin F1a values in plasma after treatment with placebo and Pycnogenol® relative to pre-treatment values

Duration of treatment	Medication	N	Percent (median)	Percentile		p _{Bon}
				25%	75%	
1 month	Placebo	30	3.07	1.70	6.27	<0.01
	Pycnogenol®	28	9.02	4.54	18.69	
2 months	Placebo	30	7.38	2.83	11.85	<0.05
	Pycnogenol®	28	11.88	7.53	21.83	
3 months	Placebo	30	8.04	2.23	12.63	<0.05
	Pycnogenol®	28	11.80	8.82	23.00	

Mann-Whitney Rank Sum Test with Bonferoni correction: *: p_{Bon} < 0.05, **: p_{Bon} < 0.01.

Table 5

Angiotensin II decrease in plasma after treatment with placebo and Pycnogenol® relative to pre-treatment values

Duration of treatment	Medication	N	Percent (median)	Percentile		p _{Bon}
				25%	75%	
1 month	Placebo	30	−7.95	−11.79	−4.97	ns
	Pycnogenol®	28	−6.16	−8.81	−3.47	
2 months	Placebo	30	−12.12	−24.23	−6.69	<0.05
	Pycnogenol®	28	−7.77	−13.70	−4.16	
3 months	Placebo	30	−18.96	−32.77	−10.58	<0.05
	Pycnogenol®	28	−9.81	−15.25	−5.71	

Mann-Whitney Rank Sum Test with Bonferoni correction: *: p_{Bon} < 0.05, **: p_{Bon} < 0.01.

Endothelin-1, angiotensin II, and 6 keto-prostaglandin F1 levels were quantified in plasma by radioimmunoassays from Beijing Huaying Biological Technology Company. Nitrogen monoxide was analyzed using a colorimetric assay for nitrite/nitrate from Nanjing Jiancheng Biological Technology Company.

Statistical analysis was done using Sigmasat 2,03 software. Results were tested for normal distribution with Kologmorov-Smirnov-test. Statistical significance was tested with Mann-Whitney Rank Sum test.

Results

Supplementation with Pycnogenol® was able to reduce the dose of nifedipine considerably (Table 2).

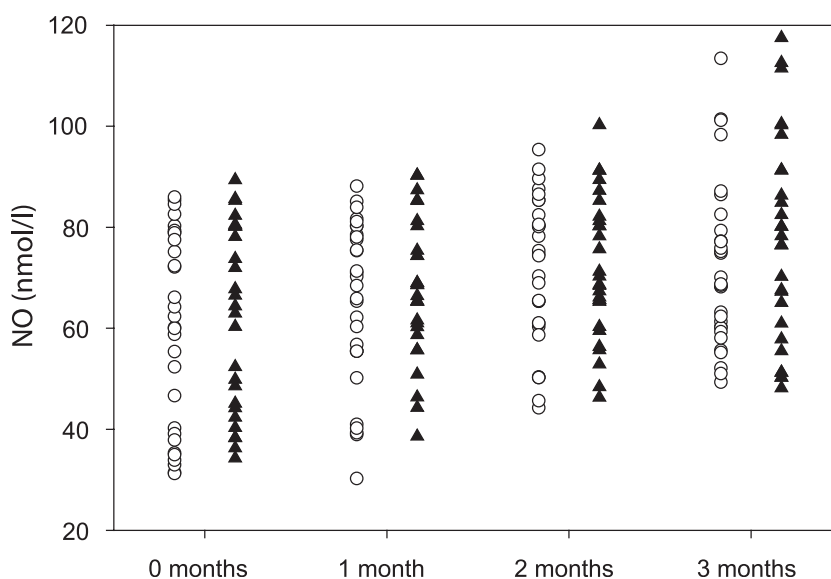


Fig. 2. Nitric oxide values in plasma after treatment with placebo (white circles) and Pycnogenol® (black triangles).

Table 6

The percentage of changes of NO concentrations in plasma after treatment with placebo and Pycnogenol® relative to pre-treatment values

Duration of treatment	Medication	N	Percent (median)	Percentile		p _{Bon}
				25%	75 %	
1 month	Placebo	30	1.70	– 4.51	24.97	ns
	Pycnogenol®	28	5.60	– 9.45	16.46	
2 months	Placebo	30	11.07	1.37	25.37	ns
	Pycnogenol®	28	10.46	– 7.04	23.96	
3 months	Placebo	30	10.50	– 2.07	29.11	ns
	Pycnogenol®	28	11.75	– 1.44	32.08	

Mann-Whitney Rank Sum Test with Bonferoni correction: *: p_{Bon} < 0.05, **: p_{Bon} < 0.01.

Most of the patients had normal blood pressures at the end of the 12 weeks under treatment with 10 mg nifedipine and 100 mg Pycnogenol®. In the placebo group, only 4 patients attained normal blood pressures with a dose of 10 mg nifedipine. The differences in nifedipine dosing between the Pycnogenol® and placebo groups were highly significant ($p > 0.001$).

Endothelin-1 (Et-1) was measured in plasma in monthly intervals (Fig. 1). A large interindividual variation of Et-1 values was observed for the placebo group, while the data for the Pycnogenol® group showed less scattering. The decrease of Et-1 levels under Pycnogenol® treatment was pronounced and statistically significant compared to placebo (Table 3) after 1 and 2 months; however, the obvious decrease after 3 months failed to reach significance because of the large variation of data.

The concentrations of 6 keto-prostaglandin F1 (PG), the metabolite of prostacyclin, increased in both groups. While large interindividual variations were observed, the increase of the PG values in the Pycnogenol® group was significantly higher compared to the placebo group (Table 4).

Values for angiotensin II decreased in both groups, the decrease being larger for the placebo compared to the Pycnogenol® group. That difference became statistically significant after 2 months of treatment (Table 5).

NO concentrations in plasma increased over the treatment period in both groups (Fig. 2). Data after intake of Pycnogenol® showed a trend towards higher values, however, differences in comparison to placebo failed to reach statistical significance (Table 6).

No statistically significant differences were observed between both groups in heart rate, electrolytes in blood, creatinine and blood urea nitrogen (data not shown).

Unwanted effects were reported by both groups of patients. These were mild and transient and none of the patients left the study because of these unwanted effects. Gastrointestinal problems, nausea, dizziness, headache and sleepiness had been reported. The difference in the rate of side effects in Pycnogenol® group (39%) and in placebo group (27%) was not statistically significant.

Discussion

Control of blood pressure was achieved with a lower dose of nifedipine in those patients receiving Pycnogenol® supplementation. In the placebo group the mean dose was 21,5 mg, while in the

Pycnogenol® group 15 mg was sufficient to lower the blood pressure to normal. This finding is consistent with the observation from a double-blind, placebo controlled, cross-over study showing a normalization of blood pressure in mild hypertensive patients (Hosseini et al., 2001) after supplementation with Pycnogenol®.

This significant antihypertensive effect could be based on several mechanisms. It is certainly an important finding that the supplementation with Pycnogenol® significantly lowered endothelin-levels by 20% as compared to placebo controls in which endothelin concentrations decreased by only 4%. Because endothelin is the most potent endothelial-derived vasoconstrictor, the lowering of endothelin concentrations should contribute to the antihypertensive effect.

Vascular tonus is also regulated by the vasodilator prostacyclin. Concentrations of the metabolite of prostacyclin, the 6 keto-prostaglandin-F1a, were significantly increased under Pycnogenol® supplementation as compared to placebo.

Pycnogenol® may shift the balance between vasodilatation and vasoconstriction towards vasodilator predominance. The observed decrease of thromboxane B2 levels after intake of Pycnogenol® in smokers (Araghi Nicknam et al., 1999) and in non-smoking, hypertensive patients (Hosseini et al., 2001) also points in that direction. Thromboxane B2 is the stable metabolite of thromboxane, another agent involved in vasoconstriction.

A fundamental process in the regulation of vascular tone is the production of NO. NO, the endothelial dependent relaxing factor, counteracts the effects of adrenaline and noradrenaline. Pycnogenol® stimulates the production of NO in isolated aortic rings (Fitzpatrick et al., 1998) and inhibits adrenaline- or noradrenaline-induced vasoconstriction. In the present study, NO concentrations increased during the treatment period with Pycnogenol® to a greater extent than in the placebo group; however, the difference between both groups failed to reach statistical significance (Table 6).

Remarkably, Et-1 concentrations in the placebo group also decreased after the pretreatment period although the decrease was not as great as after Pycnogenol® supplementation. Also, other factors indicating improved endothelial function, such as increased levels of NO and PG, were observed in the placebo group. The explanation for the pronounced placebo effect could be found in the firmer control of the patient and increasing compliance to medication and medical advice, so that treatment efficacy was greatly improved just by their inclusion in the study.

Based on an in vitro study by Blazso et al., (1996) that showed an inhibition of the angiotensin converting enzyme, it was expected that Pycnogenol® would decrease angiotensin II levels in blood during treatment. This was indeed observed, but, unexpectedly, the decrease in the placebo group was significantly greater. These results do not support the view that the clinically relevant action of Pycnogenol® is as an ACE inhibitor.

Other biological effects of Pycnogenol® may contribute to the protection of the vascular system of the hypertensive patients. The excellent ability of Pycnogenol® to inactivate free radicals was demonstrated in several systems (Rohdewald, 2002), e.g., the oxygen radical absorbance capacity of the blood of obese volunteers is indeed higher after intake of Pycnogenol® (Devaraj et al., 2002). Therefore, it can be expected that endothelial dysfunction resulting from oxidative stress could be reduced after supplementation with Pycnogenol®. Oxidative stress and LDL levels contribute to the development of atherosclerosis. Pycnogenol® influences positively the atherosclerotic index of lipoproteins by lowering LDL and increasing HDL concentrations in obese volunteers (Devaraj et al., 2002; Koch, 2002).

Taking into account that the concentration of adhesion factors is also lowered by Pycnogenol® (Peng et al., 2000), one may consider that the inflammatory processes, participating in impaired endothelial

function are lessened by the combination of antioxidative and anti-inflammatory factors (Rohdewald, 2002). Finally, the inhibition of platelet aggregation by Pycnogenol[®], which has been demonstrated in smokers (Pütter et al., 1999), is very important in blocking another detrimental factor associated with the risk of arteriosclerosis and thrombus formation. To summarize, all of these favorable effects of Pycnogenol[®] in addition to lowering blood pressure, may reduce the risk factors for patients with hypertension.

Conclusion

Our study demonstrated that supplementation with the French maritime pine bark extract Pycnogenol[®] was able to reduce significantly the dose of the calcium antagonist nifedipine in treating patients with hypertension. On average more than 5 mg nifedipine could be spared by the simultaneous administration of 100 mg Pycnogenol[®]. While the clinical significance of this drug sparing effect may not seem important based only on the effect upon blood pressure, the data obtained regarding mediators affecting the endothelium point to a more general beneficial effect of the administration of Pycnogenol[®].

The simultaneous decrease of the vasoconstrictor, endothelin-1, and increase of the vasodilator, prostacyclin, measured indirectly by analyzing 6-keto-prostaglandin F1, demonstrated a statistically significant improvement of endothelial function. Taking into account its ability to inhibit platelet aggregation and its diverse antioxidative and anti-inflammatory actions, (Rohdewald, 2002), we suggest that Pycnogenol[®] offers a broad spectrum of protection for the patient with hypertension.

References

- Araghi Nicknam, M., Hosseini, S., Larson, D., Rohdewald, P., Watson, R.R., 1999. Pine bark extract reduces platelet aggregation. *Integrative Medicine* 2, 73–77.
- Blazso, G., Gaspar, R., Gábor, M., Rűve, H.-J., Rohdewald, P., 1996. ACE inhibition and hypotensive effect of procyanidins-containing extract from the bark of *Pinus pinaster* sol. *Pharmaceutical Pharmacology Letters* 6, 8–11.
- Devaraj, S., Kaul, N., Schönlau, F., Rohdewald, P., Jialal, I., 2002. Supplementation with a pine bark extract rich in polyphenols increases plasma antioxidant capacity and alters plasma lipoprotein profile. *Lipids* 37 (10), 931–934.
- Fitzpatrick, D.F., Bing, B., Rohdewald, P., 1998. Endothelium-dependent vascular effects of Pycnogenol[®]. *Journal of Cardiovascular Pharmacology* 32, 509–515.
- Hosseini, S., Jeongmin, L., Sepulveda, R.T., Rohdewald, P., Watson, R.R., 2001. A randomized, double-blind, placebo-controlled, prospective, 16-week crossover study to determine the role of Pycnogenol[®] in modifying blood pressure in mildly hypertensive patients. *Nutrition Research* 21, 1251–1260.
- Koch, R., 2002. Comparative study of Venostasin and Pycnogenol[®] in chronic venous insufficiency. *Phytotherapy Research* 16, 1–5.
- Peng, Q., Wei, Z., Lau, B.H.S., 2000. Pycnogenol[®] inhibits tumor necrosis factor-(TNF- α)-induced nuclear factor κ B activation, and adhesion molecule expression in human vascular endothelial cells. *Cellular Molecular Life Science* 57, 834–841.
- Pütter, M., Grottemeyer, K.H.M., Würthwein, G., et al., 1999. Inhibition of smoking-induced platelet aggregation by aspirin and Pycnogenol[®]. *Thrombosis Research* 95, 155–161.
- Rohdewald, P., 2002. A review of the French maritime pine bark extract (Pycnogenol[®]), a herbal medication with a diverse clinical pharmacology. *International Journal of Clinical Pharmacology and Therapeutics* 40 (4), 158–168.