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Pomegranate juice consumption inhibits serum angiotensin converting enzyme activity and reduces systolic blood pressure

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Abstract

Consumption of pomegranate juice which is rich in tannins, possess anti-atherosclerotic properties which could be related to its potent anti-oxidative characteristics. As some antioxidants were recently shown to reduce blood pressure, we studied the effect of pomegranate juice consumption (50 ml, 1.5 mmol of total polyphenols per day, for 2 weeks) by hypertensive patients on their blood pressure and on serum angiotensin converting enzyme (ACE) activity. A 36% decrement in serum ACE activity and a 5% reduction in systolic blood pressure were noted. Similar dose-dependent inhibitory effect (31%) of pomegranate juice on serum ACE activity was observed also in vitro. As reduction in serum ACE activity, even with no decrement in blood pressure, was previously shown to attenuate atherosclerosis, pomegranate juice can offer a wide protection against cardiovascular diseases which could be related to its inhibitory effect on oxidative stress and on serum ACE activity.

1. Introduction

The pomegranate tree, which is said to have flourished in the garden of Eden, has been extensively used as a folk medicine in many cultures [1].

Content of soluble polyphenols in pomegranate juice varies within the limits of 0.2–1.0%, depending on variety and include mainly tannins, ellagic tannins, anthocyanins, catechins, gallic and ellagic acids [2,3]. We have recently shown [4] a potent anti-atherogenicity of pomegranate juice consumption in healthy humans and in atherosclerotic mice and identified tannins as the active components responsible for the anti-oxidative properties of pomegranate juice against low-density lipoprotein oxidation [4,5].

Hypertension is a known risk factor for the development of atherosclerosis and in hypertensive patients with elevated plasma rennin–angiotensin activity, a five folds increased incidence of myocardial infarction was demonstrated [6]. The production of angiotensin II from angiotensin I is inhibited by angiotensin converting enzyme (ACE) inhibitors. Angiotensin converting enzyme (ACE, EC 3.4.15.1, dipeptidyl carboxypeptidase) is a glycoprotein peptidyldipeptide hydrolase that cleaves histidylleucine dipeptide from angiotensin I (a relatively inactive decapeptide), forming the potent vasoconstrictor angiotensin II.

Clinical studies have demonstrated that ACE inhibitors significantly reduced the morbidity and mortality in patients with myocardial infarction and the incidence of recurrent myocardial infarction and ischemic events in patients with coronary artery disease, even in the absence of blood pressure lowering [7–9]. The mechanism by which ACE inhibitors affect atherosclerosis is not well understood, but it has been postulated that these agents may have multiple effects, including blood pressure lowering, anti-proliferative effect on vascular cells, inhibitory effect on platelet aggregation and inhibition of lipid peroxidation [10,11]. Data from animal experiments indicate that ACE inhibitors can attenuate the development of atherosclerosis across a wide range of species including hamsters, watanabe heritable hyperlipidemic rabbits, cholesterol-fed cynomologus monkeys and minipigs [12–14].

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Some antioxidants such as vitamin C, vitamin E, β-caroteone and coenzyme Q were recently shown to possess hypotensive properties [15–17].

As pomegranate juice possesses very potent anti-oxidative properties, we questioned the effect of pomegranate juice on blood pressure and on ACE activity.

2. Methods

Seven hypertensive males and three females with mean blood pressure levels of $155 \pm 7/83 \pm 7$ mmHg were studied. Their age ranged between 62 and 77 and they were all nonsmokers. Two of the patients were diabetics (serum glucose > 126 mg/dl) and two were hyperlipidemic (serum cholesterol > 240 mg/dl).

Eight patients were on ACE inhibitors therapy (enalapril or ramipril) and two patients were on calcium channel blockers.

Serum angiotensin II converting enzyme activity was determined in serum samples obtained before and 2 weeks after pomegranate juice consumption (50 ml contained 1.5 mmol of total polyphenols per day) by the hypertensive patients. The patients adherence to this protocol was confirmed by serum total polyphenols analysis [18]. We have also studied the in vitro effect of pomegranate juice on serum ACE activity. Serum ACE activity was measured by a commercial kit (Sigma Co. Ltd, St. Louis, MO).

This spectrophotometric method utilizes the synthetic tripeptide substrate *N*-[3-(2-furyl)acryloyl]-L-phenylalanylglcylglcine (FAPGG). FAPGG is hydrolyzed by ACE to furylacryloylphenylalanine (FAP) and glycylglycine. Hydrolysis of FAPGG results in a decreased absorbency at 340 nm. Serum ACE activity is determined by comparing the sample reaction rate to that obtained with an appropriate ACE calibrator.

3. Results and discussion

In seven out of ten studied hypertensive patients, serum ACE activity was significantly decreased by 36% after 2 weeks of pomegranate juice consumption (Fig. 1(A)). The inhibitory effect of pomegranate juice consumption on serum ACE activity (Fig. 1(A)) can be secondary to the antioxidant properties of pomegranate juice [3,4], and/or it may be related to a direct effect of the juice active compounds on serum ACE activity.

To assess a possible direct effect of pomegranate juice on serum ACE activity, increasing concentrations of pomegranate juice were added to human serum and incubated for 15 min at 37°C. A pomegranate juice dose-dependent inhibitory effect, up to 31%, on serum ACE activity was obtained (Fig. 1(B)). This effect may be secondary to the ability of pomegranate juice associated antioxidants, such as complexed tannin [3,4], to inhibit ACE activity.

The inhibitory effect of pomegranate juice consumption on serum ACE activity may have also resulted from a direct interaction of pomegranate juice constituent with serum ACE. The effect of pomegranate juice consumption on blood pressure, though minimal (5%), showed a low, but significant ($P < 0.05$) reduction in the systolic blood pressure (Table 1).

The inhibitory effect of pomegranate juice consumption may have contributed to some extent, to the reduction in blood pressure but as there is no significant correlation between the reduction in serum ACE activity and blood pressure, the reduction in ACE activity may not be the major contributor for lowering blood pressure.

Recently, the potential of antioxidant therapy to affect blood pressure in hypertensive patients was stud-

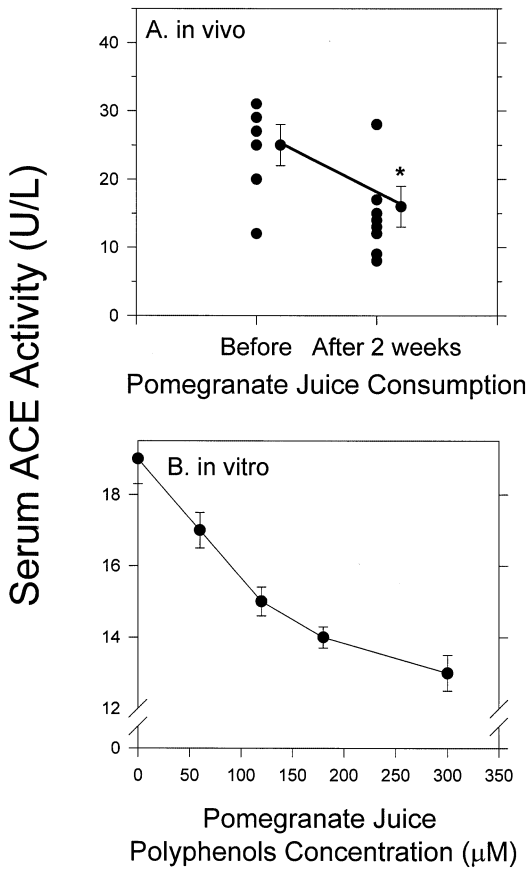

Fig. 1. The effect of pomegranate juice consumption (50 ml containing 1.5 mmol of total polyphenols per day) for 2 weeks (individual variability and mean values (A) and the in vitro dose effect of pomegranate juice (B) on serum angiotensin II converting enzyme activity.

Table 1
The effect of pomegranate juice consumption by hypertensive patients on their blood pressure

Patients	Blood pressure (mmHg)	
	Before (0 time)	After pomegranate juice consumption (2 weeks)
1	161/73	153/80
2	162/83	163/80
3	168/75	160/80
4	157/92	143/87
5	150/80	153/85
6	146/80	141/80
7	140/92	140/90
8	153/70	130/70
9	158/90	130/82
10	160/90	150/85
Mean ± S.D.	155 ± 7/83 ± 7	147 ± 10/82 ± 5

ied [19]. As reactive oxygen species contribute to endothelium-dependent contraction and to increased vascular resistance, antioxidants can possibly restore endothelial function and hence decrease blood pressure [19]. Increased serum ACE activity is associated with enhanced susceptibility to lipid peroxidation [11] and hence, the inhibitory effect of pomegranate juice on serum ACE activity can further contribute to an antioxidant environment and attenuated atherosclerotic risk. Hypotensive effects of dietary antioxidants may be secondary to increasing availability of the vasodilator nitric oxide, which can affect ACE activity [16]. An additional possibility for the effect of pomegranate juice consumption to reduce serum ACE activity can be possibly related to an effect on cytochrome P-450 enzymes as these enzyme breakdown drugs such as ACE inhibitors. Inactivation of P-450 3A4 was indeed shown to occur by grapefruit juice [20] and such an effect could have resulted in the presence of a relatively high serum levels of the ACE inhibitor drug, and hence in lowering serum ACE activity. Unlike grapefruit juice, however, pomegranate juice has only minimal inhibitory effect on P-450 3A4 activity (M. Aviram, preliminary observation), and thus, such an effect may not be relevant to pomegranate juice.

Reduction in serum ACE activity with the potent ACE inhibitor ramipril, even with no reduction in blood pressure, possesses anti-atheogenic properties in mice [21], and it is also associated with reduced mortality in cardiovascular patients [22].

We thus conclude that the significant inhibitory effect of pomegranate juice on serum ACE activity and the minor attenuation in blood pressure in hypertensive patients, in addition to its potent inhibitory effect on lipid peroxidation [4], suggests that pomegranate juice consumption can offer a wide protection against cardiovascular diseases.

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