

# Effects of pomegranate juice on cardiometabolic risk factors and biomarkers of oxidative stress and inflammation in hemodialysis patients, a crossover controlled trial

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## Abstract

**Background:** Pomegranate has antioxidant, cardioprotective, and anti-inflammatory properties. We questioned if pomegranate juice (PJ) benefits lipid profile and oxidative and inflammatory biomarkers of hemodialysis patients.

**Methods:** The study had a crossover design. Hemodialysis patients (n=41) were divided into two groups: PJ-treated group who received 100 ml natural PJ immediately after their dialysis session three times a week and the control group who received the usual care. After 8 weeks, a 4-week washout period was established and afterwards the role of the groups was exchanged. Lipid profile, blood pressure, and oxidative and inflammatory biomarkers were measured before and after each sequence. Comparisons between the two conditions were performed by ANCOVA with adjustments for potential confounders.

**Results:** Based on results of intention-to-treat analysis, triglycerides were decreased in PJ condition and increased in the control. Conversely, high-density lipoprotein (HDL)-cholesterol was increased in PJ and decreased in control. There was a significant between-condition difference for both triglycerides and HDL-cholesterol ( $P < 0.001$ ). Total and low-density lipoprotein (LDL)-cholesterol did not significantly change in either condition. Systolic and diastolic blood pressure significantly decreased in PJ condition and caused a significant difference between two conditions ( $P < 0.001$ ). Total antioxidant capacity increased in PJ condition ( $P < 0.001$ ) and decreased in control ( $P < 0.001$ ). Conversely, malondialdehyde and interleukin-6 decreased in PJ ( $P < 0.001$ ) and increased in the control ( $P \leq 0.001$ ). The changes of these biomarkers were significantly different between two conditions.

**Conclusions:** In conclusion, 8-week PJ consumption showed beneficial effects on blood pressure, serum triglycerides, HDL cholesterol, oxidative stress, and inflammation in hemodialysis patients. The trial was registered at Iranian Registry of Clinical Trials (ID number: IRCT 2016070428797N1).

**Keywords:** Pomegranate, hemodialysis, hypertension, oxidative stress, inflammation, lipid profile.

## Background

Chronic kidney disease (CKD) is a long-term kidney disease characterized by gradual loss of kidney function [1]. End-stage renal disease (ESRD) occurs in the last stages of CKD, when kidneys lose most

of their function in filtering blood from wastes [1]. Although accumulation of fluid, toxins, and wastes in the body is the primary dilemma in ESRD, the patients are implicated in comorbidities such as cardiovascular diseases [2,3], mainly as a result of blood lipid disorders [1,4], hypertension [5], oxidative stress [3], and inflammation [6].

Lipid profile abnormalities in CKD generally include elevated triglycerides and reduced levels of high-density lipoprotein (HDL) cholesterol with either no remarkable change or a reduction in low-density lipoprotein (LDL) cholesterol [7]. This is especially important because compared to total and LDL cholesterol, low HDL cholesterol and elevated triglyceride concentrations predict more strongly cardiovascular disease [7]. These lipid abnormalities are likely due to post-translational modification of apolipoproteins, especially B100, by glycation, oxidation, and carbamylation (the last one resulted from spontaneous decomposition of urea) which leads to prolonged delipidation and thus appearance of small-dense LDL particles and highly atherogenic chylomicron and very low-density lipoprotein remnants [7].

High blood pressure is both a trigger and a consequence of CKD [8]. High blood pressure in CKD may be the result of fluid retention, altered sodium homeostasis, and consequently activation of renin-angiotensin-aldosterone system [8]. Also, oxidative stress which comes along with CKD may promote endothelial dysfunction through suppression of nitric oxide bioavailability.

CKD is also a case of oxidative stress and inflammation. Oxidative stress may be considered as a tool for prediction of morbidity and mortality in CKD patients [9,10]. CKD patients have low levels of antioxidants, such as vitamin C and glutathione, and high concentration of pro-oxidants because of increased production of reactive oxygen and nitrogen species by a variety of means, for instance, damaged nephrons, uremic toxins, and bio-incompatibility of dialysis membranes and solutions [11,12]. On the other hand, oxidative stress and inflammation go hand in hand; it is not only oxidative stress that acts as a trigger for inflammatory response but inflammation in turn recruits phagocytes into the injured region, leading to production of oxygen radicals [13].

Hemodialysis is the last strategy for the treatment and survival of ESRD patients when they are on waiting list for kidney transplantation. While the procedure attenuates blood pressure, it has no effect

on lipid profile, but also exacerbates severity of oxidative stress and inflammation [12]. Thus, consumption of foods with broad biological advantageous may be beneficial for ESRD patients. Pomegranate (*Punica granatum* L.) is an ancient fruit, cultivated all around the world. Due to extensive health benefits, pomegranate has been the focus of many *in vitro*, animal, and human studies [14]. The beneficial effects of pomegranate are attributed to polyphenols which are phytochemicals with great antioxidant, anticarcinogenic, anti-diabetic, cardioprotective, and anti-inflammatory capabilities [15].

Very few available studies on the effect of pomegranate in hemodialysis patients found conflicting results for cardiovascular risk factors and no effect for markers of oxidative stress and inflammation [16-18]. The controversy or negative results may be due to the time of pomegranate administration which has been mostly before dialysis session when polyphenols is quickly excreted during dialysis. Therefore, in the current study we examined the effect of postdialysis administration of pomegranate juice (PJ) on lipid profile and biomarkers of oxidative stress and inflammation in hemodialysis patients.

## Methods

### **Study design**

The trial was preformed according to a crossover design in spring and summer 2016. Hemodialysis patients were selected from dialysis centers in Namazi and Shahid Faghihi hospitals of Shiraz, Iran. A sample size of 19 was determined based on results provided by previous investigators [19], a type 1 error of 5%, power of 20%, and considering a 10% drop-out rate.

Inclusion criteria were as follows: ESRD patients on dialysis treatment, dialysis 3 times a week for at least 3 months, an age range of 18 to 65 years, life expectancy more than 1 year, serum potassium level of less than 6 mEq/L, and permission of the physician in charge of the patient. Patients were not included if they were involved in serious diseases such as malignancy, other organs' failure, taking antioxidant supplements such as vitamin E and vitamin C over the month before the study, iron sucrose (Venofer) usage at least two weeks before the study, malnutrition that required nutritional support, and hospitalization for > 5 days per month. Our primary protocol was to exclude the patients

if they lost inclusion criteria, missed PJ consumption for > 3 days a month, changed hypertension and hyperlipidemia medications, did not follow dietary recommendations, and hospitalized during the intervention. However, none of the patients was excluded according to these criteria.

The study was approved by the Ethics Committee of Shiraz University of Medical Sciences (approval number. IR.SUMS.REC.1394.210), and registered at Iranian Registry of Clinical Trials (ID number: IRCT 2016070428797N1). Study protocol was explained and written informed consent was obtained from all participants.

### **Intervention**

The study was performed in a crossover design with two 8-week sequences separated by a 4-week washout period. At the beginning, the participants were divided into two groups: PJ-treated group (n = 22) who consumed 100 g natural PJ after their dialysis session three times a week and the control group (n = 19) who received the usual care. After the 8-week intervention, there was a 4-week washout course and afterwards the second sequence of the study was started in which the role of the groups was interchanged. The Ethical Committee of our institution did not permit having a control treatment because 1) hemodialysis patients have restrictions for consumption of liquids; 2) it may not be ethically approved to ask hemodialysis patients to drink a placebo beverage when a beneficial effect for which could not be substantiated; 3) making a control drink for PJ required addition of artificial colors and pomegranate flavor which could be harmful for ESRD patients; and 4) in contrast to similar studies on hemodialysis patients, we administered PJ after dialysis session, thus it was crucial to avoid administering unnecessary food items. Because hemodialysis patients spend hours in the dialysis unit, to prevent contamination between intervention and control groups, we recruited the participants from two hospitals and allocated each of the intervention and control groups to a hospital at a time.

Pomegranate was from Rabab variety cultivated in Neyriz, Shiraz. Pomegranates were purchased from Neyriz pomegranate farms in autumn and stored at -20 °C until the time of the intervention. During the intervention, each week some pomegranates were thawed, crushed, and squeezed, and the juice was collected in 500 ml bottles. Thus, the juice was prepared fresh and distributed amongst

the patients in PJ condition every week. The patients were asked to keep the juice in a refrigerator until the time of use. One hundred milliliters measuring cups were provided for the participants and they were asked to drink 100 ml PJ three times a week after their dialysis session. In order to ensure consumption of entire amount of PJ, each week 100 ml extra PJ was given to the patients just in case if other family members wanted to taste it.

During the study, each patient was under supervision and care of a nephrologist and medical care team. Blood pressure was measured before and after PJ consumption in order to ensure that PJ would not cause hypotension. For all of the patients, weight maintenance diets were prescribed and the amount of fruit and vegetables that each patient could consume was explained. Diets were set in such a way that people in both groups received equal amounts of fruit and fruit juice. A list of high-potassium fruits was given to the patients and they were asked to abstain from high-potassium fruits on days of PJ consumption. The patients were also recommended to exclude pomegranate and PJ consumption throughout the intervention except for the PJ which was administered.

### **Measurements**

Total antioxidant capacity and malondialdehyde were considered as the primary and interleukin-6 was measured as the secondary outcome. All parameters were measured at baseline, the end of the first sequence, and before and after the second sequence of the intervention. For weight and body composition, measurements were performed immediately after the dialysis session [20]. Weight was measured by a digital scale (AM-2018) and body composition was determined by bioelectric impedance analyzer (BIA) (InBody, Biospace Co., South Korea). Blood pressure was measured in predialysis state twice with at least 5 min interval by a digital barometer (Microlife BP A200 AFIB). The mean of two measurements was considered as the subject's blood pressure.

Blood samples were collected after 12-h fasting. All biochemical variables were measured in serum. Serum lipid profile was analyzed by an auto-analyzer (BT 1500, Biotechnica Instruments, Italy) using specific kits (Pars-Azmun, Iran). Malondialdehyde (MDA) was quantitated by thiobarbituric acid reactive substances (TBARS) method [21]. The method principle is based on the reaction of lipid peroxidation products in serum samples with thiobarbituric acid in the presence of trichloroacetic

acid, butylated hydroxytoluene, hydrochloric acid, and sodium dodecyl sulfate. The reaction mixture was then heated for 30 min at 95 °C and the formed chromogen is read at 532 nm. Interleukin-6 (IL-6) (Diacalone, France) and serum total antioxidant capacity (TAC) (Biocore diagnostics, Hamburg, Germany) were determined by colorimetric assay using commercially available kits.

Three-day dietary recall (two weekdays and one weekend day) was used to assess dietary intake and monitor dietary compliance. Dietary intakes were analyzed by Nutritionist IV software (First Databank, San Bruno, CA, USA) modified for Iranian foods. Mini nutritional assessment (MNA) was used as a tool for detecting malnutrition in hospitalized young and middle-aged adults [22].

### **Statistical analyses**

Data were analyzed using SPSS software (version 22; IBM, Armonk, USA). Normality of data distribution was checked by visual examination of the histogram curve of data frequencies. In the case of abnormality, log-transformed data was used for determination of statistical difference between treatment conditions. The intention-to-treat (ITT) approach was used for the analysis. Comparisons between the two conditions were performed by ANCOVA (treatment × time interaction) with age and gender as the covariates. P value < 0.05 was considered statistically significant.

### **Results**

After assessing 182 individuals, 57 patients who met the described inclusion criteria were found and 41 patients accepted to participate in the study. The patients were assigned to PJ (n=22) and control (n=19) groups for the first sequence. Due to stomach discomfort, one patient from PJ group could not continue PJ intervention and was excluded. In the second sequence, one person from the PJ group deceased and another moved away. Therefore, 38 and 40 patients completed the 8-week PJ and control conditions, respectively (Figure 1). However, the analysis was performed based on ITT approach, with 41 and 40 patients being in the PJ and control conditions, respectively. Table 1 indicates baseline characteristics of participants. The age range of participants was between 24 and 65 years, and 61% of them were males. They had hemodialysis duration ranging from 1 to 15 years. PJ consumption did not affect anthropometric measures including weight, BMI, waist circumference, skeletal muscle and fat mass; no significant difference was either observed between the two

conditions (Table 2). However, total body water increased significantly in control condition and the changes were significantly different between the conditions ( $P=0.009$ ). Mini nutritional assessment significantly decreased in PJ condition ( $P<0.001$ ) and increased in the control ( $P=0.003$ ); the difference between the two conditions was also significant ( $P<0.001$ ).

Systolic and diastolic blood pressure significantly decreased in PJ condition ( $P<0.001$ ). Systolic blood pressure increased significantly in control condition and the difference between PJ and control conditions for both systolic and diastolic blood pressure was significant ( $P<0.001$ ). Triglycerides decreased in PJ condition ( $P<0.001$ ) and increased in the control ( $P=0.007$ ). In contrast, HDL cholesterol increased in PJ condition ( $P<0.001$ ) and decreased in the control ( $P=0.001$ ). There was a significant between-condition difference for both triglycerides and HDL cholesterol ( $P<0.001$ ). Total and LDL cholesterol did not significantly change in either condition. Similarly, no significant alteration in AST and ALT was observed within or between the conditions.

Total antioxidant capacity increased in PJ condition ( $P<0.001$ ) and decreased in the control ( $P<0.001$ ). Conversely, malondialdehyde and IL-6 decreased in PJ condition ( $P<0.001$ ) and increased in the control ( $P\leq 0.001$ ). The changes of these parameters were significantly different between the two conditions.

Serum sodium, calcium, and phosphate did not change significantly during the study in either condition but potassium levels increased in PJ condition ( $P=0.01$ ) with no significant difference between control and PJ conditions. Dietary intakes did not differ between PJ and control conditions (Table 3).

## Discussion

Our study showed that 8-week PJ consumption (100 ml/day three times a week) may have beneficial effects on blood pressure, serum triglycerides and HDL cholesterol of hemodialysis patients. PJ may also have antioxidant and anti-inflammatory effect as it increased total antioxidant capacity and decreased malondialdehyde and IL-6. In the amounts consumed, PJ did not have adverse effects on potassium levels.

Hypertension is one of medical problems in hemodialysis patients mostly originated from fluid

overload [20]. Conversely, reduction of total body water in CKD could be associated with better blood pressure control [23]. In the current study, a significant difference was observed between PJ and control conditions in the amount of total body water. This difference occurred as a result of non-significant decrease in total body water of patients in PJ condition and a significant increase in total body water of patients in the control. These alterations in total body water were concordant with changes in blood pressure in each condition. Although a cause-effect relationship could not be predicated from results presented here, other investigators have shown that reduction in total body water may improve blood pressure in hemodialysis patients [23].

Previous studies have not examined the effect of pomegranate or polyphenols on total body water content of hemodialysis patients but the blood pressure-lowering effect of PJ has been reported in previous trials on both normotensive [24] and hypertensive individuals [25], patients with metabolic syndrome [26], and hemodialysis patients [18]. A meta-analysis of randomized controlled trials confirmed this effect independent of duration or dose of PJ [27]. The beneficial effect of pomegranate on blood pressure may be due to improved nitric oxide bioavailability through scavenging reactive oxygen species [28] as well as enhanced activity and expression of endothelial nitric oxide synthase [29]. In addition, pomegranate may promote vasorelaxation by inhibition of angiotensin converting enzyme [30]. These effects are attributed to phenolic compounds of pomegranate [31].

In contrast to blood pressure, evidence regarding the effect of pomegranate on blood lipids of hemodialysis patients is rather controversial. One study confirmed our results for serum triglycerides and HDL cholesterol [18] but two other investigations reported no effect of either pomegranate polyphenols [16] or PJ [17] on these lipids. Some of the controversy over the results of different studies may be explained by baseline values of triglycerides and HDL cholesterol in participants which likely need to be in unfavorable levels in order for pomegranate to exert its benefit. Also, consumption of pomegranate before dialysis session may reduce its beneficial effect because pomegranate polyphenols pass through dialysis membrane and are discarded in the dialysate [32,33]. Similarly, the lack of PJ effect on total and LDL cholesterol in our study may be partly due to relatively normal levels of these lipids in the participants as other studies on diabetes patients with elevated

blood lipids showed promising results [34,35]. The lipid-lowering and anti-atherogenic properties of pomegranate may relate to a number of mechanisms such as regulating expression of genes involved in lipogenesis and fatty acid oxidation in the liver [36], increasing expression and activity of paraoxonase-1, the antioxidant enzyme in HDL particles that prevents LDL oxidation [37], and suppression of oxidized LDL degradation and of cholesterol synthesis in macrophages [38]. In agreement with us, investigations have reported pomegranate antioxidant potential as evidenced by decreased malondialdehyde [34,35,39,40] and increased serum total antioxidant capacity [41]. Likewise, the anti-inflammatory effect of pomegranate has been shown by reduction of C reactive protein [26,35] and IL-6 [35,41], the latter as an inflammatory biomarker with highest predictive value for ESRD outcome [42]. Once again, pomegranate may not exert these effects in non-oxidative stress conditions [25,43] or if administered in pre-dialysis states [17]. Also, such beneficial effects have not been observed in high doses of pomegranate and long intervention lengths [16]. The beneficial effects of pomegranate are attributed to polyphenols [15]. Polyphenols in pomegranate are mostly from anthocyanin subcategory [44] that have great antioxidant and anti-inflammatory properties [15].

Based on the results of MNA, nutritional status was significantly improved in PJ and worsened in control condition. Although less common and valid than subjective global assessment and malnutrition inflammation score methods [45], MNA has been successfully used for assessment of nutritional status in CKD patients [45-48]. A cross-sectional study on hemodialysis patients documented positive correlations between MNA and serum albumin, fat and muscle mass, and BMI and a negative correlation with C reactive protein levels [47]. Nevertheless, no consistency was found in this study between MNA scores and anthropometric characteristics which could be because of relatively short duration of the intervention that did not allow remarkable change in anthropometric measures. In addition, the incongruence between MNA and fat and muscle mass results may be due to low accuracy of BIA in the diagnosis of malnutrition [49].

There were limitations with this study. Due to dietary restrictions and ethnic issues related to hemodialysis patients, as explained in detail in the methods, we could not consider a control

treatment for the control arm. Also, BIA may not have shown body composition with sufficient accuracy. More complex techniques such as dual-energy X-ray absorptiometry may determine body composition with more precision. Nonetheless, the study had a crossover design which means that all participants experienced both PJ and control condition, thus reducing risk of selection bias. Also, administration of PJ after dialysis session allows longer cessation of PJ polyphenols in the circulation which enhances their effectiveness. Natural PJ is preferred to pomegranate supplements because it can be embedded in the regular diet more easily.

## Conclusions

In conclusion, post-dialysis consumption of 100 ml/day PJ three times a week for 8 weeks had beneficial effects on blood pressure, serum triglycerides and HDL cholesterol, and oxidative and inflammatory condition of hemodialysis patients without having adverse effect on potassium levels. However, these results may not be generalized to hemodialysis patients from other geographical regions, mainly due to the difference in pomegranate phytochemical composition.

## Abbreviations

ALT: alanine transaminase; ANCOVA: analysis of covariance; AST: aspartate aminotransferase; BIA: bioelectric impedance analyzer; BMI: body mass index; CKD: chronic kidney disease; ESRD: end-stage renal disease; HDL: high-density lipoprotein; IL-6: interleukin-6; LDL: low-density lipoprotein; SD: standard deviation; MDA: malondialdehyde; MNA: mini nutritional assessment; PJ: pomegranate juice; TAC: total antioxidant capacity; TBARS: thiobarbituric acid reactive substance.

## Declarations

### **Acknowledgement**

Not applicable.

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### **Availability of data and materials**

The datasets generated during the current study are available from the corresponding author on reasonable request.

### **Authors' contributions**

RBB and MA contributed to the conception and design of the study. RBB prepared the pomegranate juice and gave it to the participants. RBB and ZE recruited the participants, gave dietary recommendations, conducted the trial, and collected the data. MMS medically supervised the patients. RBB and MA analyzed the data. RBB and ZE drafted the manuscript and MA revised it. All authors read and approved the final version of the manuscript.

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### **Ethics approval and consent to participate**

All participants gave written informed consent. The study was performed according to guidelines of 1964 Declaration of Helsinki and its later amendments. The trial was approved by the Ethics Committee of Shiraz University of Medical Sciences (approval number: IR.SUMS.REC.1394.210) and registered in the Iranian Registry of Clinical Trials (IRCT 2016070428797N1) at <https://en.irct.ir/trial/23270?revision=23270>.

## Consent for publication

Not applicable.

## Competing interests

The authors declare no conflict of interest.

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## Tables

**Table 1 - Baseline characteristics of the participants**

Characteristics	Mean $\pm$ SD or n (%)
Age (y)	47.8 $\pm$ 13.3
Males (n)	25 (61.0)
Weight (kg)	65.7 $\pm$ 15.5
Body mass index (kg/m <sup>2</sup> )	23.9 $\pm$ 4.8
Mini nutritional assessment (points)	15.2 $\pm$ 2.8
Systolic blood pressure (mm Hg)	135.9 $\pm$ 8.3
Diastolic blood pressure (mm Hg)	98.0 $\pm$ 9.5
Triglycerides (mg/dl)	154.4 $\pm$ 75.1
Total cholesterol (mg/dl)	150.1 $\pm$ 38.0
LDL cholesterol (mg/dl)	76.1 $\pm$ 25.7
HDL cholesterol (mg/dl)	43.3 $\pm$ 10.3
AST (U/L)	13.6 $\pm$ 7.8
ALT (U/L)	11.1 $\pm$ 7.1

Data are expressed as either mean  $\pm$  standard deviation

(SD) or number and percentage. n=41. Abbreviations:

ALT, alanine aminotransferase; AST, aspartate

aminotransferase; HDL, high-density lipoprotein; LDL,

low-density lipoprotein.

Table 2 - The effect of 8 weeks' PJ consumption on anthropometric, blood pressure, and biochemical measurements (values are the sum of both sequences)<sup>1</sup>

	Baseline	Week 8	Difference (95% CI)	P v (tir)
Weight (kg)				
PJ condition	65.65 $\pm$ 15.67	65.78 $\pm$ 15.41	0.13 (-0.11, 0.38)	0
Control condition	65.65 $\pm$ 15.67	65.65 $\pm$ 15.45	0.00 (-0.22, 0.22)	0
Body mass index (kg/m <sup>2</sup> )				
PJ condition	23.88 $\pm$ 4.86	23.93 $\pm$ 4.77	0.05 (-0.03, 0.14)	0
Control condition	23.88 $\pm$ 4.86	23.88 $\pm$ 4.77	0.00 (-0.08, 0.08)	
Waist circumference (cm)				

PJ condition	85.63 ± 13.50	86.17 ± 14.10	0.54 (-0.33, 1.41)	0
Control condition	84.18 ± 15.11	83.85 ± 14.88	-0.33 (-1.10, 0.45)	0
Fat mass (kg)				
PJ condition	17.81 ± 11.76	18.02 ± 11.73	0.21 (-0.19, 0.61)	0
Control condition	18.35 ± 12.16	17.21 ± 11.84	-1.15 (-2.76, 0.47)	0
Skeletal muscle mass (kg)				
PJ condition	26.73 ± 5.85	26.58 ± 5.63	-0.14 (-0.43, 0.14)	0
Control condition	26.67 ± 6.12	26.88 ± 6.00	0.21 (-0.13, 0.55)	0
Total body water (kg)				
PJ condition	35.83 ± 7.21	35.62 ± 6.97	-0.21 (-0.53, 0.11)	0
Control condition	35.72 ± 7.49	36.11 ± 7.07	0.38 (0.03, 0.74)	0
Mini nutritional assessment (points)				
PJ condition	15.68 ± 2.79	14.18 ± 2.65	-1.50 (-1.90, -1.10)	<0
Control condition	14.88 ± 2.88	15.50 ± 2.65	0.63 (0.23, 1.02)	0.1
Systolic blood pressure (mm Hg)				
PJ condition	135.65 ± 8.47	128.69 ± 6.11	-6.97 (-4.88, -9.05)	<0
Control condition	135.95 ± 8.27	136.93 ± 7.46	0.97 (0.11, 1.84)	0
Diastolic blood pressure (mm Hg)				
PJ condition	97.73 ± 9.95	90.84 ± 6.86	-6.88 (-9.08, -4.68)	<0
Control condition	98.20 ± 9.34	99.18 ± 8.88	0.98 (-0.29, 2.24)	0
Triglycerides (mg/dL)				
PJ condition	157.22 ± 69.79	125.32 ± 58.24	-31.90 (-18.77, -45.04)	<0
Control condition	150.77 ± 81.59	171.67 ± 102.59	20.90 (6.16, 35.64)	0.1
Total cholesterol (mg/dL)				
PJ condition	155.07 ± 38.97	149.82 ± 36.46	-5.26 (-16.96, 6.44)	0
Control condition	144.25 ± 36.80	145.85 ± 30.45	1.60 (-6.87, 10.07)	0
LDL cholesterol (mg/dL)				
PJ condition	80.20 ± 25.96	77.77 ± 23.86	-2.43 (-9.16, 4.30)	0
Control condition	71.82 ± 25.38	75.80 ± 28.60	3.97 (-1.92, 9.87)	0
HDL cholesterol (mg/dL)				
PJ condition	42.40 ± 9.52	52.33 ± 18.46	9.93 (5.38, 14.48)	<0
Control condition	44.60 ± 10.88	39.20 ± 11.01	-5.40 (-2.32, -8.48)	0.1
AST (U/L)				
PJ condition	13.82 ± 8.95	12.70 ± 6.39	-1.13 (-3.60, 1.35)	0
Control condition	13.35 ± 6.74	14.25 ± 7.89	0.90 (-1.79, 3.59)	0
ALT (U/L)				
PJ condition	10.62 ± 7.07	9.55 ± 5.92	-1.08 (-3.07, 0.91)	0
Control condition	11.57 ± 7.32	11.87 ± 7.68	0.30 (-1.81, 2.41)	0
Total antioxidant capacity (mmol/L)				

PJ condition	0.40 ± 0.08	0.49 ± 0.11	0.09 (0.05, 0.13)	<0
Control condition	0.47 ± 0.08	0.40 ± 0.08	-0.07 (-0.04, -0.10)	<0
Malondialdehyde (µmol/L)				
PJ condition	0.88 ± 0.01	0.77 ± 0.01	-0.11 (-0.07, -0.15)	<0
Control condition	0.86 ± 0.01	0.91 ± 0.01	0.05 (0.02, 0.08)	0.1
Interleukin-6 (ng/L)				
PJ condition	3.00 ± 1.48	2.09 ± 1.25	-0.90 (-0.56, -1.24)	<0
Control condition	2.18 ± 1.17	3.05 ± 1.60	0.87 (0.53, 1.22)	<0
Sodium (mEq/L)				
PJ condition	140.4 ± 4.90	141.1 ± 5.86	0.73 (-1.90, 3.37)	0
Control condition	140.8 ± 5.66	142.1 ± 5.50	1.32 (-1.39, 4.04)	0
Potassium (mEq/L)				
PJ condition	5.56 ± 0.28	5.69 ± 0.30	0.13 (0.03, 0.23)	0
Control condition	5.54 ± 0.25	5.57 ± 0.30	0.03 (-0.08, 1.37)	0
Calcium (mg/dL)				
PJ condition	7.95 ± 0.41	7.97 ± 0.50	0.02 (-0.20, 0.25)	0
Control condition	8.05 ± 0.47	8.07 ± 0.58	0.02 (-0.22, 0.26)	0
Phosphate (mg/dL)				
PJ condition	4.90 ± 0.45	4.93 ± 0.44	0.03 (-0.16, 0.23)	0
Control condition	4.92 ± 0.40	5.00 ± 0.38	0.08 (-0.09, 0.26)	0

<sup>1</sup> Data are means ± SD. n=41 in PJ condition and n=40 in control condition. <sup>2</sup> P value assessed by paired samples t test. <sup>3</sup> Comparisons between the two conditions were performed by ANCOVA (treatment × time interaction) with age, gender, and baseline values as the covariate. Abbreviations: ALT, alanine aminotransferase; AST, aspartate aminotransferase; HDL, high-density lipoprotein; ITT, intention-to-treat analysis; LDL, low-density lipoprotein.

Table 3 - Dietary intakes during the study periods<sup>1</sup>

	PJ condition	Control condition	P value <sup>2</sup>
Energy (kcal/day)	1394 ± 366.5	1310 ± 328.0	0.30
Carbohydrate (g/day)	203.8 ± 60.25	187.5 ± 51.98	0.21
Protein (g/day)	58.49 ± 16.29	56.32 ± 12.64	0.52
Fat (g/day)	37.89 ± 9.73	36.92 ± 9.98	0.67
Saturated fatty acids (g/day)	13.78 ± 4.43	13.93 ± 5.34	0.90
Monounsaturated fatty acids (g/day)	12.61 ± 3.05	12.24 ± 2.94	0.59
Polyunsaturated fatty acids (g/day)	7.23 ± 2.09	6.85 ± 1.60	0.37
Cholesterol (g/day)	218.57 ± 76.39	226.14 ± 69.09	0.65
Fiber (g/day)	8.54 ± 3.27	7.43 ± 2.99	0.13
Vitamin E (mg/day)	2.25 ± 0.93	2.24 ± 1.17	0.94
Vitamin C (mg/day)	35.30 ± 16.24	31.67 ± 10.68	0.26
Carotenoids (µg/day)	409.0 ± 542.1	524.3 ± 517.0	0.35
Sodium (mg/day)	1475 ± 517.5	1452 ± 565.6	0.86
Potassium (mg/day)	1486 ± 399.2	1361 ± 320.8	0.14
Calcium (mg/day)	574.2 ± 168.5	620.4 ± 198.3	0.28
Phosphorus (mg/day)	796.7 ± 189.2	780.7 ± 173.9	0.70

<sup>1</sup> Data are means ± SD. n=41 in PJ condition and n=40 in control condition. <sup>2</sup> P value assessed by independent samples t test. ITT, intention-to-treat analysis.

## Figures

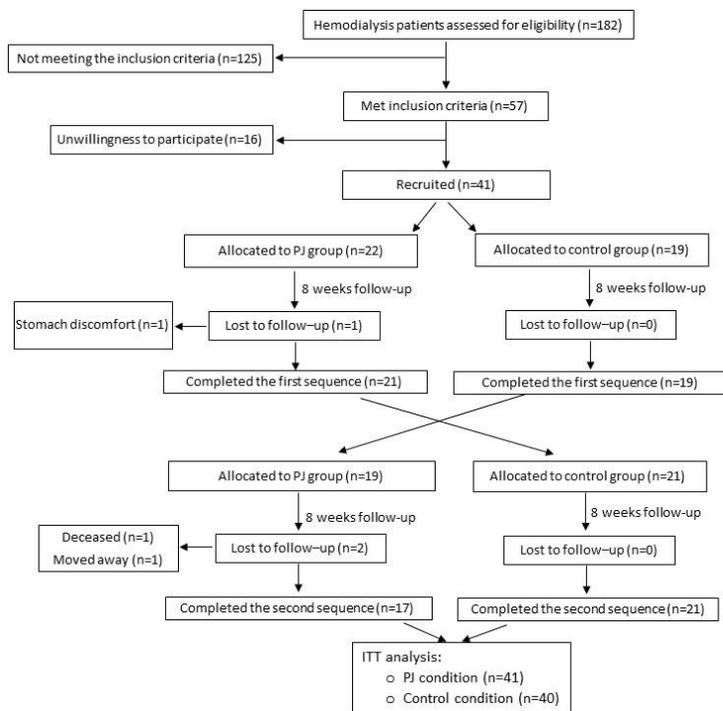


Figure 1

Flowchart of the trial - ITT, intention-to-treat.

## Supplementary Files

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