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The Effectiveness and Safety of Pomegranate (*Punica granatum* L.) Juice Powder in Patients with Diabetic Nephropathy, a Double-Blinded Randomized Clinical Trial

Elnaz Faridi[1], Pouya Faridi[1], Mehrdad Vosoughi[2], Ramin Ansari[3], Amirreza Dehghanian[4], Jamshid Roozbeh[5], Mohammad Mehdi Zarshenas[1]*

[1]Department of Phytopharmaceuticals (Traditional Pharmacy), School of Pharmacy, Shiraz University of Medical Sciences, Shiraz, Iran
[2]Department of Community Oral Health, School of Dentistry, Shiraz University of Medical Sciences, Shiraz, Iran
[3]Department of Clinical Pharmacy, School of Pharmacy, Tehran University of Medical Sciences, Tehran, Iran
[4]Department of Pathology, School of Medicine, Shiraz University of Medical Sciences, Shiraz, Iran
[5]Department of Internal Medicine, School of Medicine, Shiraz University of Medical Sciences, Shiraz, Iran

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Abstract

Diabetic nephropathy affects more than 40 % of diabetic patients and is the major cause of death in this population. Current medications, including angiotensin-converting enzyme inhibitors (ACEIs), angiotensin receptor blockers (ARBs), statins, sodium-glucose cotransporter-2 inhibitors, calcitriol, etc., represent limitations both in terms of maximum efficacy and adverse effects. Animal studies exist regarding the nephroprotective effects of *Punica granatum* L. (Pomegranate) extract. The present study aims to assess the safety and efficacy of pomegranate juice in patients with diabetic nephropathy. Freeze-dried pomegranate juice powder was purchased from Shaanxi Tianxingjian Co. and filled in 500 ± 5 mg capsules. The intervention group (n= 23) received 4 capsules of pomegranate daily for 8 weeks; while the placebo group (n= 23) received identical-looking capsules similarly. All patients received maximum tolerated doses of ACEI /ARB, Statin, and fixed doses of Empagliflozin for the last 6 months. Fasting blood sugar, serum creatinine, blood urea nitrogen, aspartate transaminase, alanine transaminase, total cholesterol, high-density lipoprotein cholesterol, low-density lipoprotein cholesterol, urine albumin, and urine albumin-creatinine ratio were measured at baseline and the end of the 4th and 8th weeks. Among all of the investigated parameters, urine albumin-creatinine ratio, and urine albumin were significantly reduced ($p < 0.001$) in the intervention group after 4 and 8 weeks as compared with the baseline and the placebo group. Other investigated parameters represent no differences in none of the groups. No adverse reaction was reported during the study. Previous studies suggest that pomegranate possesses anti-inflammatory properties by inhibiting interleukin (IL)-1, IL-6, IL-8, tumor necrosis factor-α, and transforming growth factor-β. This is the first clinical trial study investigating the efficacy of pomegranate in diabetic nephropathy, suggesting a promising role for this formulation as a supplement for patients receiving standard treatment, since it remarkably reduced the indicators of diabetic nephropathy.

Keywords: Diabetic nephropathy; *Punica granatum*; Clinical trial; Urine albumin

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*Corresponding Author: Mohammad Mehdi Zarshenas
Department of Phytopharmaceuticals (Traditional Pharmacy), School of Pharmacy, Shiraz University of Medical Science, Shiraz, Iran
Email: zarm@sums.ac.ir

Introduction

Diabetic nephropathy, as a progressive disease, affects more than 40% of diabetic patients worldwide. Moreover, it is the major cause of death in this population [1,2]. Reduced Glomerular Filtration Rate (GFR), sustained albuminuria, hypertension, and edema might result from diabetic nephropathy.

Without management, 25-40 % of the patients move toward End-stage Kidney Disease (ESKD). Due to the chronic nature of the disease, there has been no definite treatment for the disease till now [3-5].

Reactive Oxygen Species (ROS) play a critical role in the disruption of renal vasculature. An increase in blood glucose and free fatty acids potentiates the production of ROS, advanced glycation end-products, and an inflammatory cascade. Inflammatory cytokines (including tumor necrosis factor (TNF)-α, interleukin (IL)-6, IL-1, vasoactive peptides, adhesive molecules, and growth factors are all involved in the pathophysiology of the disease, leading to glomerulosclerosis, mesangial expansion, interstitial fibrosis, tubular atrophy, and damage to the vasculature basement membrane [6-8].

Current medication for diabetic nephropathy includes strict regulation of blood glucose, hypertension, dyslipidemia, smoking cession, weight loss, and medications reducing proteinuria including Angiotensin-Receptor Blockers (ARBs), Angiotensin Converting-Enzyme Inhibitors (ACEIs), Sodium-Glucose Transport Protein 2 (SGLT2), Glucagon-like peptide 1 (GLP-1) receptor agonists, Aldosterone receptor antagonist, Statins, and Calcitriol [9]. Due to the limited effects of current medications, as well as adverse reactions (e.g., electrolyte imbalance, hypotension, and myalgia) that limit the tolerable dose, there is a need for other medications/supplements to control the disease.

Due to the global trend toward complementary and alternative medicine, various studies have previously investigated the nephroprotective effects of herbal medications in animal models of diabetic nephropathy. *Panax quinquefolius* L., *Brassica oleracea* L., *Eugenia jambolana* Lam., *Paeonia albiflora* Pallas, *Silybum marianum* L., *Zingiber officinale* Roscoe, and *Cinnamomum verum* J.Presl are herbal medicines showed promising results both as prevention and management of diabetic nephropathy in animal model mainly to the effects on regulation of blood glucose, antioxidant effects and due to the flavonoid content [10,11].

Based on the pharmacopeias of Traditional Persian Medicine (TPM), including the *Canon of Medicine*, *Punica granatum* L. (pomegranate) is recommended as a nephroprotective agent [12]. Previous studies represent the anti-inflammatory and anti-hyperglycemic effects of the plant as the main protective approaches for the disease [13].

To facilitate consumption and increase patient compliance, uniformity of preparation, and stability of the final product [14], we considered preparing a solid dosage form of the aqueous extract of *P. granatum*.

The present study aims to clinically evaluate the efficacy and safety of *P. granatum* juice powder in patients with diabetic nephropathy.

Materials and Methods

Ethical approval

Before study enrollment, all patients provided written informed consent. The trial was conducted under the Declaration of Helsinki guidelines and reported following the Consolidated Standards of Reporting Trials (CONSORT) statement [15]. The study was approved by the Ethics Committee of Shiraz University of Medical Sciences (Registration number: IR.SUMS.REC.1394.132) and registered in the IRCT clinical trials registry (IRCT2015121725574N1).

Formulation preparation

Freeze-dried pomegranate juice powder was purchased from Shaanxi Tianxingjian Co., China (Batch number: LXSL140203). Roasted starch was used to prepare the placebo. Both the drug and placebo were filled in identical-looking zero-size capsules (500 ± 5 mg).

Standardization of the formulation

Using the spectrophotometry method, total phenol and total flavonoid contents were determined based on gallic acid and quercetin, respectively. The quantification of the polyphenolic content of the extract was carried out using the Folin-Ciocalteu reagent (Sigma Aldrich- USA), considering the standard method [16]. The methanolic solutions of gallic acid (Merck- Germany) with concentrations of 3.125, 6.25, 12.5, 25, 50, and 100 mg/mL were prepared as standard [17]. After the preparation of the calibration curve, the absorption was reported at 765 nm at 20˚C. Phenolic content was reported to be equivalent to milligrams of gallic acid per 1 gram of pomegranate juice powder.

The total amount of flavonoids was quantified using Aluminum chloride colorimetry (A modified Dowd method) [18]. Different concentrations of quercetin (Sigma-Aldrich) (20, 40, 60, 80 mg/lit) were prepared in methanol (Merck- Germany). Then, 2.5 mL of 2% Aluminum chloride (Sigma Aldrich- USA) was added to each, and the absorption was read at 415 nm (PG instrument T90 spectrophotometer- Germany) after 10 minutes. The pomegranate juice was prepared in a concentration of 1000 mg/lit in methanol (Merck-Germany), and the absorption was read similarly. The total flavonoid content was reported as mg quercetin

equivalent/g of the pomegranate juice powder.

Patients and setting

This study is a single-center, randomized, double-blind, placebo-controlled clinical trial conducted at the Shahid Motahari Clinic of Shiraz University of Medical Sciences (SUMS). Participant recruitment occurred between October 2021 and March 2022.

Patients aged between 25 to 75 years old with type 2 diabetes with moderately increased albuminuria (urine albumin-creatinine ratio (UACR) of 30–300 mg/g (based on the three separated urine samples collected over two weeks before the study initiation), referring to the outpatient clinic of the Shahid Motahari were included in the study after confirmation by a nephrologist.

All the patients have a history of receiving fixed and maximum tolerated doses of ACEI, or ARB, and statin for at least 6 months before entering the study. Since SGLT2 inhibitors affect proteinuria and it is recommended as a drug of choice for diabetic patients with proteinuria, all the patients received a fixed dose (10 mg daily) of Empagliflozin for at least the previous 6 months before entering the study.

Patients with a history of kidney transplantation, those on dialysis, stage III or IV of heart failure, type 1 diabetes, active infection, concurrent use of corticosteroids, non-dihydropyridine calcium channel blockers, aldosterone receptor antagonists, GLP-1 receptor agonists, pregnant and lactating women, concurrent autoimmune disease or malignancy, myocardial infarction, transient ischemic attack within 6 months before the beginning of the study, stroke, those with non-diabetic renal failure, and those with previous hypersensitivity reaction to pomegranate were excluded from the study.

A computer-generated statistical program determined the sample size, calculating a minimum of 18 patients per group based on $\alpha = 0.05$, $\beta = 0.1$, and a desired power of 90%. Patients were randomized to treatment groups using a block randomization method (block number = 4). To maintain allocation concealment, sequentially numbered, opaque, sealed envelopes were used. A third party, the clinic secretary, opened the envelopes after the patient consented to assign treatment groups.

Patients in the intervention group received 2 capsules (each 500 ± 5 mg) of pomegranate juice after lunch and 2 capsules after dinner for 2 months. The placebo group received identical-looking capsules in the same manner.

The demographic data of the patients were recorded at the beginning, including age, gender, duration of the disease, weight, and height. The body mass index (BMI) and eGFR (using the Cockcroft-Gault formula) of the patients were calculated. Fating blood sugar (FBS), serum creatinine, blood urea nitrogen (BUN), aspartate transaminase (AST), alanine transaminase (ALT), total cholesterol, high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), urine albumin, and UACR were measured at baseline and the end of the 4th and 8th weeks. Patients were clinically visited by a specialist in nephrology at baseline and the end of the 4th and 8th weeks. Adverse effects of the therapy, if any, were also recorded during the study interval.

Statistical analyses

Data analysis was conducted using IBM SPSS Statistics Version 20. Continuous variables are expressed as mean ± standard deviation. Statistical significance was set at p value < 0.05. The Kolmogorov-Smirnov test was employed to assess the normality of data distribution. The Wilcoxon test was used to compare the mean values between the placebo and drug groups. Independent t-tests were utilized to evaluate between-group differences at various time points; while repeated-measures ANOVA was employed to assess the trend of variable changes over time.

Results

Total Phenol and total Flavonoid content

According to the line equation obtained from the standard gallic acid curve ($y = 0.0041x - 0.0229$, $R^2 = 0.9965$), the average concentration of phenol content in 1000 mg/Lit solution of pomegranate juice was equal to 29.11±0.22 mg. In other words, in each 1000 mg of pomegranate juice powder, about 30 mg of phenol content exists (30 mg gallic acid equivalent/g dried extract). Based on the line equation obtained from the standard quercetin curve ($y = 0.0279x - 0.0095$, $R^2 = 0.9992$), the average concentration of flavonoid content in 1000 mg/lit solution of pomegranate juice was equal to 4.93±0.16. In other words, in each 1000 mg of pomegranate juice powder, about 5 mg of flavonoid content exists (5 mg quercetin equivalent/g dried extract).

Clinical trial study

Of the 74 patients initially screened, 28 were excluded due to not meeting the inclusion criteria. 46 eligible patients were randomized into two groups of 23 patients each. Two patients from each group were lost to follow-up during the study period. Finally, 21 patients completed the study in each group (Figure 1).

The intervention group consisted of 10 (47.61%) men and 12 (57.14%) women, and the placebo group consisted of 11 (53.39%) men and 9 (42.86%) women. There was no significant difference between groups in terms of sex ($p = 0.324$).

The mean age of patients in the intervention group

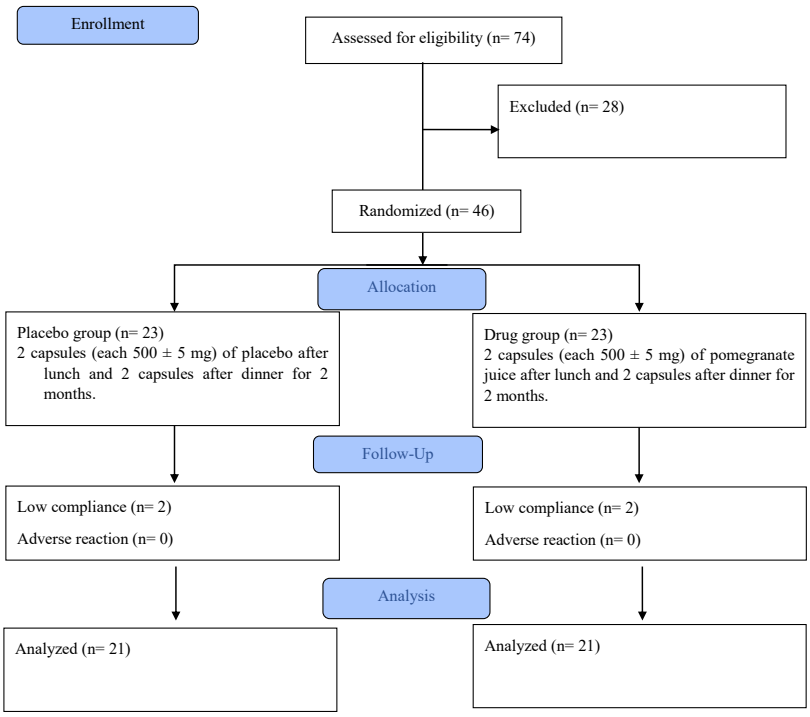

Figure 1. Consort flowchart

was 51.82 ± 2.50 years; while the mean age in the placebo group was 56.28 ± 2.24 years. A comparison of the two groups revealed no statistically significant difference in age (p = 0.201).

Similar results were obtained for the duration of diabetes, BMI, and eGFR while comparing the groups at baseline (p =0.378, 0.621, and 0.088 respectively). As shown in Table 1 none of the laboratory parameters including FBS (p =0.883), AST (p =0.559), ALT (p =0.822), LDL-C (p =0.556), HDL-C (p =0.114), Total Cholesterol (p =0.766), Triglyceride (p =0.065), Serum Cr (p =0.284), BUN (p =0.467), UACR (p =0.752), and Urine Albumin (p =0.980) represent significant differences between groups at baseline.

As has been shown in table 2, results of the within-group statistical analysis suggest no remarkable differences in terms of FBS, BMI, AST, ALT, HDL-C, LDL-C, Total Cholesterol, Triglyceride, Serum Cr, BUN, and eGFR in none of the groups during the study interval at 4th week and 8th week (p > 0.05). Results of the intragroup analysis represent similar results and suggest no statistically significant differences between groups in both the 4th week and 8th week. Consequently, none of the groups showed any remarkable changes during the study interval.

Unlike other studied parameters, while considering UACR and urine albumin, the interaction of time and group (Group × Time) is statistically meaningful (p < 0.001). In this scenario, the separate p value for the group type and the p value for time are not reliable. Hence, the subgroup analysis was conducted using an independent t-test to evaluate the effect of the drug and placebo at baseline, 4th week, and 8th week separately.

There were no statistically significant differences between the two groups at baseline in terms of UACR and urine albumin (p = 0.752, 0.362). Furthermore, no significant differences were observed at the 4-week (p = 0.182, 0.162) and 8-week (p = 0.106, 0.188) follow-up assessments.

Using a one-sample repeated measure ANOVA test while considering the type of group in the 8th week, significant differences were observed for the intervention group (p = 0.006, 0.001), but not the placebo group (p = 0.070, 0.112). So, the reduction in UACR and urine albumin is remarkable just in the intervention group during the study interval. Moreover, remarkable differences were observed in UACR and urine albumin in the intervention group while considering the differences between baseline and the 4th week (p = 0.027, 0.011), and while considering the differences between baseline and the 8th week (p = 0.019, 0.026).

No serious adverse events or clinically significant side effects were observed during the study.

Discussion

P. granatum has been traditionally used for diabetes,

Table 1. Demographic data of patients before entrance to the clinical study

Variables	Drug group Mean± SD (n=21)	Placebo group Mean± SD (n=21)	*p* value
Sex. No (%)			
Male	10 (47.61)	11 (52.39)	0.324
Female	12 (57.14)	9 (42.86)	
Age (Year)	51.82 ± 2.50	56.28 ± 2.24	0.201
Duration of diabetes (Years)	5.86 ± 3.63	5.32 ± 3.44	0.378
Urine Albumin (mg/dL)	6.25 ± 1.26	6.14 ±1.88	0.752
UACR (mg/g)	92.68± 32.46	85.66± 28.26	0.980
FBS (mg/dL)	166.00 ± 18.44	161.83 ± 21.44	0.883
BMI (kg/m2)	27.98 ± 1.15	27.15 ± 1.21	0.621
ALT (U/lit)	22.41 ± 1.71	23.22 ± 3.37	0.822
AST (U/lit)	21.205 ±1.19	23.50 ± 4.06	0.559
LDL-C (mg/dL)	90.064 ± 6.07	84.35 ± 7.60	0.556
HDL-C(mg/dL)	48.82 ± 3.19	42.08 ± 2.44	0.114
Triglyceride (mg/dL)	186.64 ± 19.04	138.78 ± 15.35	0.065
Total cholesterol (mg/dL)	169.36 ± 7.55	165.28 ± 11.92	0.766
Serum Cr (mg/dL)	1.22 ± 0.13	1.42 ± 0.14	0.284
BUN (mg/dL)	21.94 ± 2.95	25.24 ± 3.39	0.467
eGFR (mL/min)	61.57 ± 9.65	69.85 ±6.93	0.088

Cr = Serum Creatinine, BUN = Blood Urea Nitrogen, BMI=Body Mass Index, eGFR = Estimated Glomerular Filtration Rate, ALT=Alanine aminotransferase, AST=Aspartate aminotransferase, LDL-C=Low-Density Lipoprotein cholesterol, HDL-C=High-Density Lipoprotein cholesterol, UACR= Urine Albumin-Creatinine Ratio

inflammatory disease, diarrhea, and bowel infection in TPM [19]. Anti-hyperglycemic, antioxidant, anti-inflammatory, and antitumor effects of the plants are mainly attributed to the suppression of the inflammatory mediators/pathways including vascular cell adhesion protein 1 (VCAM-1), angiotensin II activity, TNF-α, TGF-β, IL-1, IL-6, IL-8, P38 mitogen-activated protein kinases (MAPK), Monocyte chemoattractant protein (MCP)-1, and c-Jun N-terminal kinases (JNK) pathway mainly due to the flavonoid and polyphenol content of the plant [19-21].

Most of the medicinal plants previously evaluated as nephroprotective agent works through the regulation of blood glucose and improving glucose tolerance, including *Terminalia chebula* Retz., *Olea europaea* L., *Capsicum frutescens* L., *Salvia officinalis* L., *Citrullus colocynthis* (L.) Schrad., and *S. marianum*. Moreover, various polyherbal formulations have been introduced in traditional medicines for diabetic nephropathy [22]. This is the first study investigating the effects of pomegranate in diabetic nephropathy patients.

Results regarding the effect of *P. granatum* on blood glucose are controversial. Results of a study suggest pomegranate juice did not affect blood glucose [13]; while another study suggests 30% reduction in blood glucose in a group of mice fed with pomegranate seed [23]. In our study, pomegranate juice reduced blood glucose, although it was statistically insignificant. Similar to our study, the results of a previous study regarding the effect of pomegranate on BMI in patients with hyperlipidemia suggest an insignificant reduction in this population. Although there are pieces of evidence toward the effect of pomegranate juice (40 g concentrated pomegranate juice for 8 weeks) on total cholesterol and LDL-C in patients with type 2 diabetes and concurrent hyperlipidemia [24,25], our study suggests no remarkable effect on the lipid profile, including LDL-C and total cholesterol. HDL-C represents no change in a previous study, similar to our results. Triglyceride was reduced insignificantly in our study in line with previous studies, which suggest unchanged triglyceride following pomegranate administration [24,25]. While considering the results of systematic reviews, there is a report regarding the insignificant effects of pomegranate supplementation on FBS, glycosylated hemoglobin (HbA1c), insulin level, BMI, triglyceride, HDL-C, and LDL-C in patients with type 2 diabetes [26]. On the other hand, another systematic review suggests that pomegranate can remarkably decrease glycemia, body weight, triglyceride, cholesterol, and LDL-C levels [27]. These variations might result from the diversity of the included study population, design, pomegranate dosing, and duration.

Although the effect of pomegranate on AST and ALT has not been clinically investigated to date in patients

Table 2. Comparison of the Mean±SD of the investigated parameters between the drug and placebo groups during the study period.

Variables	Baseline (Mean± SD)	4th week (Mean± SD)	8th weeks (Mean± SD)	*p* value (Group × Time)	*p* value (Group)	*p* value (Time)
FBS (mg/dL)						
• Drug	166.00 ± 18.44	161.41 ± 18.15	162.41 ± 16.29	0.512	0.508	0.396
• Placebo	161.83 ± 21.44	152.94 ± 16.24	133.33 ± 13.34			
BMI (kg/m^2)						
• Drug	27.98 ± 1.15	28.05 ± 1.11	28.18 ± 1.14	0.337	0.571	0.560
• Placebo	27.15 ± 1.21	27.08 ± 1.26	27.10 ± 1.25			
ALT (U/lit)						
• Drug	22.41 ± 1.71	21.64 ± 1.61	22.80 ± 1.97	0.474	0.542	0.516
• Placebo	23.22 ± 3.37	25.28 ± 3.85	25.50 ± 4.80			
AST (U/lit)						
• Drug	21.21 ± 1.19	21.85 ± 1.90	23.99 ± 2.74	0.523	0.656	0.224
• Placebo	23.50 ± 4.06	24.78 ± 4.44	24.72 ± 4.83			
LDL-C (mg/dL)						
• Drug	90.06 ± 6.07	95.54 ± 8.39	96.85 ± 7.07	0.809	0.433	0.165
• Placebo	84.35 ± 7.60	87.99 ± 6.06	87.41 ± 6.70			
HDL-C (mg/dL)						
• Drug	48.82 ± 3.19	45.48 ± 2.99	49.41 ± 2.58	0.172	0.218	0.563
• Placebo	42.08 ± 2.44	44.44 ± 2.84	43.61 ± 2.74			
Triglyceride (mg/dL)						
• Drug	186.64 ± 19.04	182.52 ± 16.53	176.59 ± 15.97	0.725	0.063	0.738
• Placebo	138.78 ± 15.35	144.56 ± 15.36	139.67 ± 11.70			
Total Cholesterol (mg/dL)						
• Drug	169.36 ± 7.55	174.00 ± 11.23	172.32 ± 9.17	0.403	0.397	0.833
• Placebo	165.28 ± 11.92	159.44 ± 8.50	157.78 ± 10.01			
Serum Cr (mg/dL)						
• Drug	1.22 ± 0.13	1.25 ± 0.14	1.31 ± 0.15	0.547	0.432	0.198
• Placebo	1.42 ± 0.14	1.40 ± 0.13	1.45 ± 0.18			
BUN (mg/dL)						
• Drug	21.95 ± 2.95	22.40 ± 3.04	23.66 ± 3.62	0.636	0.581	0.159
• Placebo	25.24 ± 3.39	23.78 ± 2.35	26.37 ± 3.62			
eGFR (mL/min)						
• Drug	91.57 ± 9.65	95.04 ± 12.76	87.31 ± 9.50	0.449	0.126	0.487
• Placebo	69.85 ± 6.93	71.31 ± 7.45	71.48 ± 7.69			
Urine Albumin (mg/dL)						
• Drug						
• Placebo	6.25 ± 1.26	4.91 ± 0.79	4.22 ± 0.65	0.001	0.362	0.162
	6.14 ± 1.88	5.48 ± 1.11	5.41 ± 0.72			
UACR (mg/g)						
• Drug	92.68 ± 32.46	55.26 ± 22.13	46.24 ± 20.12	0.001	0.752	0.182
• Placebo	85.66 ± 28.26	81.66 ± 26.24	82.12 ± 24.12			

FBS = Fasting Blood Sugar, BMI = Body Mass Index, ALT = Alanine aminotransferase, AST = Aspartate aminotransferase, LDL-C= Low-Density Lipoprotein cholesterol, HDL-C= High-Density Lipoprotein cholesterol, Cr = Creatinine, BUN = Blood Urea Nitrogen, eGFR = Estimated Glomerular Filtration Rate, UACR = Urine Albumin-Creatinine Ratio.

with diabetic nephropathy, results of the animal studies suggest unchanged enzyme levels following pomegranate administration, similar to the results of our study [28].

It has been suggested that pomegranate flower extract might reduce BUN and Cr in mice following kidney poisoning [29, 30], but insignificant change has been reported for pomegranate juice similar to our results. On the other hand, pomegranate seeds have remarkably improved eGFR in diabetic mice [29,30]; while

the eGFR of the participants in this study remained unchanged.

Due to the limited evidence in the field, more studies with a larger population are required to fully understand the effect of pomegranate on the aforementioned parameters while considering the part of the plant used in each study.

This is the first clinical study investigating the effect of pomegranate on urine albumin and UACR. As mentioned, pomegranate juice remarkably reduced urine albumin and UACR after 4 and 8 weeks of treatment in the drug group. The results of a study suggest that pomegranate leaf has reduced proteinuria significantly in the animal model of diabetic nephropathy, mainly due to its flavonoid content [31].

The exact mechanism of how pomegranate might reduce albuminuria and UACR without affecting other parameters cannot be fully explained. Reversing vasoconstriction through improving endothelial function by the activation of nitric oxide release besides inactivation of superoxide leading to an increase in endothelial nitric oxide activity is a theory proposed for currently available medication administered for the management of the proteinuria [32].

Results of a study suggest that among patients receiving pravastatin, proteinuria was reduced without correlation to the lipid profile. Although we did not investigate the effect of pomegranate on specific biomarkers, the inhibition of renal endothelin-1, leading to decreased fibroblast proliferation and inflammatory reactions, was considered as a possible mechanism for the nephroprotective effects of the pravastatin [33]. Normalizing urinary vascular endothelial growth factor (VEGF) levels was proposed as another possible mechanism involved in reducing proteinuria [34].

Although concurrent hyperlipidemia can be potentially responsible for glomerular vasoconstriction through oxidation of LDL-C and triggering inflammatory cytokines [35]; while considering the baseline lipid profile of the patients in this study, it can be suggested that data are in near normal/normal ranges. This might be due to the statin the patients were receiving before entering the study. Hence, the statistically unchanged lipid profile of the patients in this study might result from reaching the target level before entering the study.

On the other hand, it could be hypothesized that the antioxidant and anti-inflammatory effects of pomegranate, resulting from its flavonoid and polyphenol content, may represent a synergistic effect besides standard treatment in this population.

Limitations

One of the limitations of this study was the variation in the maximum tolerated dose/ type of the ACEI/ ARB and statin that the patients received as a standard treatment. This maximum tolerated dose depended on the incidence of adverse reactions (hypotension, electrolyte imbalance, myalgia, rise in hepatic enzymes, etc.), which varied among patients. On the other hand, it was not ethical to deprive patients of the standard treatment, as we proposed that *P. granatum* might be effective as a supplement besides standard regimens. In contrast, most of the patients had other definite indications for ACEI/ARB and statin rather than reducing proteinuria. Another limitation of the study is the short-term duration of the intervention and follow-up period, which may affect the representation of the formulation's maximum efficacy and safety, although it has been suggested that no adverse effects have been reported for humans even when pomegranate juice has been consumed for up to 3 years [36].

Conclusion

This study confirms the effectiveness of pomegranate on two main indicators of diabetic nephropathy, including urine albumin and UACR. Moreover, no adverse reaction was reported during the study, suggesting the formulation's safety. Hence, pomegranate juice might be considered as a, herbal supplement for patients with diabetic nephropathy and those suffering from adverse reactions/minimum efficacy of the standard treatment.

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Conflict of Interests

The authors have no competing interests to declare that are relevant to the content of this article.

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