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LABORATORY STUDY

The effect of alfuzosin on renal resistive index, urinary electrolytes and β2 microglobulin levels and TGF β-1 levels of kidney tissue in rats with unilateral ureteropelvic junction obstruction

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ABSTRACT

Background: In this study, it was aimed to determine the effects of alfuzosin on experimentally generated unilateral partial ureteropelvic junction obstruction (UPO) in rats.

Materials and methods: Thirty Long–Evans rats were randomly allocated into five groups. In control group (C), nothing was performed; in group Sham (S) only laparotomy was done; in Alfuzosin group (A) only alfuzosin was administered for two weeks (10 mg/kg/day p.o.) without any surgery; in UPO group, unilateral UP junction obstruction was produced; and in the Group UPT (ureteropelvic obstruction + treatment), alfuzosin was administered for two weeks (10 mg/kg/day p.o.) in addition to UPO production. Renal pelvic anteroposterior diameters were determined with ultrasonography (USG) and renal arterial resistivity indexes by color Doppler USG. Urine was collected both at the beginning and at the end of the experiment for 24 h in all the groups and at the end of the experiment, blood samples were obtained. Blood and urine electrolytes and TGF-β1, urine density, urine β_2 microglobulin levels were determined. Renal tissue samples harvested from all of the rats were histopathologically evaluated. Results were determined using one-way ANOVA *t*-test; $p < 0.05$ was accepted as significant.

Results: Urine density in the UPT group was lower with respect to UPO group and blood electrolytes were preserved as close to normal ($p < 0.05$). In the UPT group, urine TGF-β1 and blood TGF-β1, blood β_2 microglobulin levels and histopathologic damage scores were lower compared to the UPO group ($p < 0.05$).

Conclusion: It is shown in this experimental unilateral partial UPO model that alfuzosin treatment prevents obstructive renal damage.

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Alfuzosin; experimental; β2 microglobulin; renal resistive index; TGF-β1; ureteropelvic junction obstruction

Introduction

Ureteropelvic junction obstruction (UPO) is the obstruction of urine passage from pelvis to ureter due to anatomic or functional reasons. UPO is the most common reason of childhood hydronephrosis and is seen in one out of 1250 live births. It is amongst the most frequent reasons of renal failure in children.[1]

Especially, renal tubular damage occurs in UPO cases.[2] It was shown that the number of aquaporins which ensures that the urine is concentrated in collector channels with proximal and distal tubules, sodium and urea transporter proteins decrease as a symptom of renal tubular function disorder.[3–6] Ultrasonography (USG), scintigraphic works, intravenous pyelography, and magnetic resonance are utilized for diagnosing obstructive uropathy, however measuring the levels of some invasive markers in urine has not come into routine use.[7] It is thought that transforming growth factor-β_1 (TGF-β_1), which constitutes as one of the most important elements of obstructive renal damage and causes renal function disorder, has a role in the formation of tubulointerstitial fibrosis. It is reported that angiotensin II increases TGF-β_1 expression in UPO and that the receptor inhibition from angiotensin converting enzyme or angiotensin II decreases TGF-β_1 expression and thus tubulointerstitial fibrosis.[8–10] Alfuzosin is a urea selective alfa$_1$ adrenoreceptor antagonist, and blocks α_{1a} found mainly in proximal ureter. It is commonly used in benign prostate hypertrophy in adults and relaxing the bladder neck in children. Alfuzosin, one of the

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selective alfa$_1$ blockers, has been used typically in prostate hypertrophy but also recently for inhibiting muscle contractions in order to ensure smooth passage of ureteral calculi.[11] It was shown that 10 mg/kg alfuzosin treatment in rats has an antioxidant effect, regulates nitric oxide (NO) release, and decreases the sympathomimetic effects.[12,13] Apart from the aforementioned effects, it was intended to investigate whether it has protective effects on tubules and kidneys by using it in partial UPO model for the first time in literature, due to the fact that it reduces intraluminal pressure in the ureter.

The main goal of this study is to evaluate whether alfuzosin has a protective effect in experimental unilateral chronic UPO treatment in rats with the help of some parameters such as renal arterial resistive index measurements, urine electrolyte and β_2 microglobulin levels, and histopathological renal damage. TGF-β_1 response in urine and serum in UPO, alfuzosin's effect on this response, and consequently whether it obstructs tubulo interstitial fibrosis will be examined as well.

Materials and methods

Long-Evans rats ranging in weight between 250 g and 300 g were utilized in this study. The animals were fed with standard feed and city water at static ambient temperature and humidity and in cages convenient for standard laboratory conditions with 12 h of light and 12 h of darkness. No antibiotics were used since they might have had an impact on the results due to their potential nephrotoxic effects. Surgical operation on the animals was conducted in sterile conditions and under general anesthesia with 40 mg/kg of ketamine hydrochloride (Ketalar, Eczacıbaşı, Turkey) and 10 mg/kg of xylazine hydrochloride (Alfazyne 2%, Ege Vet, Turkey) applied intramuscularly.

A total of 30 rats were used in five groups composed of six rats each. Each of these groups was treated differently as indicated below:

- Control group (group C): the group which was not operated on or medicated;
- Sham group (Group S): only conducted laparotomy on, but not given any other medication;
- Alfuzosin group (Group A): no surgical intervention, only treated with alfuzosin for two weeks;
- UPO group: nephrectomy was conducted on right kidney, and unilateral moderate partial chronic UPO was induced on the left;
- UPO and treatment group (Group UPT): nephrectomy was conducted on right kidney, and unilateral moderate partial chronic UPO was induced on the left, and then treated with alfuzosin for two weeks.

Creating experimental UPO model

A midline incision was cut in the abdominal under general anesthesia after cleaning the surgery region. No abnormal mass or pyelonephritis symptoms were observed in any of the rats which went under surgical operation. Left ureter was spotted and freed from surrounding tissues with a blunt dissection on the region close to UPB, and then a 3 cm long guard wire with a diameter of 0.35 mm was placed next to the ureter. The guide wire and the ureter were tied together with a free 7/0 prolene suture (Figure 1). It was made sure that each knot was as tight as the next one. The guide wire was gently removed from the suture to create a moderate partial UPO model. The incision was then closed with 4/0 silk.[14]

Two weeks were allotted in Group UPT for the ureteropelvic stenosis to become chronic, and then the animals were medicated with Alfuzosin (Xatral XL 10 mg, Sanofi Aventis, Turkey) administered in their drinking water for two weeks. A dose of 10 mg/kg/day of alfuzosin was preferred, an amount previously used on rats (12). Group UPD was just monitored for four weeks without any treatment.

Ultrasonic evaluation of kidney

USG evaluation was done by administering 40 mg/kg ketamine hydrochloride intramuscularly with a L54M 13-6 linear probe and Hitachi EUB-7500 USG device after trimming the fur on the animal's stomach. First, the renal dimensions and AP (anterior–posterior) diameters were measured, examined, and recorded. Afterwards, intrarenal arterial resistive index (ARI) values were measured during Doppler USG examination.[15] Doppler USG examinations were conducted before the experiment on

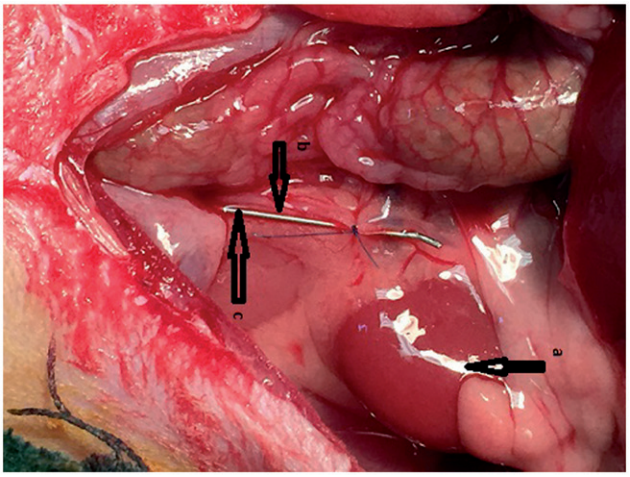


Figure 1. Creating partial pelvi-ureteric junction obstruction in rat.

the control group; before and after two weeks of laparotomy on the Sham group; before and after the completion of the treatment in the group to which only alfuzosin was administered; before the creating UPO, after moderate partial chronic UPO was created, and after the completion of the experiment on group UPO; and before creating UPO, after moderate partial chronic UPO was created, and after the completion of the treatment on Group UPT, all by the same radiologist. Each measurement was carried out three times and then their average was utilized. For Group UPO, it was made sure that there was an obstruction in ureteropelvic junction by evaluating the renal pelvis anterior–posterior diameters after the surgical operation.

Collecting urine samples and examination

The rats were held in metabolic cages for 24 h to collect their urine. Urine collection times were: twice both at the beginning and after two weeks of the experiment from Group C, twice both before and after two weeks of laparotomy from Group S, twice (both before the alfuzosin treatment and after it was finished two weeks later) from Group A, twice (both two and four weeks after UP stenosis was induced) from Group UPO, and twice (both two weeks after UPO was induced and after the two-week alfuzosin treatment was over) from Group UPT.

The densities and volumes of the urine samples were measured by using refractometer. The samples were analyzed in Gazi University Faculty of Medicine Central Biochemistry Laboratory with an autoanalyzer (Roche Cobas 8000, Indianapolis, IN) by using ready kits for Na, K, Ca, P, and Cl levels and with another (Siemens Dade Bahring) kit for β_2 microglobulin levels.

Four weeks after experimental UPO was induced, the animals were again taken into laparotomy under general anesthesia and suitable antiseptic conditions. Seeing that their left kidneys are hydronephrotic, it was acknowledged that experimental UPO was formed. Left kidneys of the animals which were sacrificed by drawing blood with intracardiac puncture were extracted together with the stenosis region. Under normal circumstances, there is no urine flow down from the stenosis region even though the kidneys are hydronephrotic; but urine outflow was seen after squeezing the kidney.

Twenty four hour urine samples were collected from groups C, S, and A which did not have UPO, and then left kidneys of the animals which were sacrificed by drawing blood with intracardiac puncture were extracted.

Blood samples drawn with intracardiac puncture were centrifuged and analyzed with an autoanalyzer (Roche Cobas 8000) by using ready kits for Na, K, Ca, P, Cl, Bun, and creatinine levels.

Extracted renal tissues were divided appropriately for histopathological examination and thus two full-thickness tissue parts were obtained. One of them was embedded in paraffin and divided into blocks following 10% formalin fixation. Four micron sections were cut from the paraffin blocks and stained with hematoxylin eosin. Ventana's automated staining device and kits were used in PAS and trichrome histochemical staining.

Glomerular structure, sclerosis, and collapse were analyzed during the histopathological examination. While proximal and distal tubules were evaluated with regards to necrosis, dilatation, and epithelial degeneration; the interstitium structure was examined with respect to edema, fibrosis, and inflammation. Vascular elements were also probed for congestion during this study.

While how many of the rats in a certain group varied according to the examination criteria during the histopathological evaluation were stated explicitly; glomerular structure and dilation of proximal and distal tubules were graded during the examination.

Remaining tissue samples were congealed in TRIzol and preserved in $-80\,^{\circ}C$, and their evaluation regarding tissue TGF-β_1 levels were planned. After conducting studies on both urine and blood, remaining urine samples and serum plasma were congealed in TRIzol and preserved in $-80\,^{\circ}C$ and their urine and blood TGF-β_1 levels were analyzed with ready to use commercial ELISA kit (Abcam TGF-β_1 Rat ELISA kit, catalog no.: ab119558, Lot: GR212552-2, UK).

Statistical analysis of the data carried out in computer environment with SPSS for Windows 20. Experimental results were reflected as mean value ± standard deviation (mean ± SD). One-way ANOVA was utilized to test the between-group variance, and the Bonferroni test was used to evaluate the within-group variance. Additionally, before and after parameters of the urine were assessed with *t*-test. $p < 0.05$ was acknowledged as statistically significant.

Results

While there is no notable difference ($p > 0.05$) regarding urine electrolytes or density (excluding the comparison between Group UPO and Group S about their repeating Na, P, and K value measurements), it was observed that urine density in Group UPO has increased significantly ($p = 0.048$) with regards to the repeating measurements within-group from before and after the experiment and that other groups did not undergo any change regarding their start and end values (Table 1).

Table 1. Weekly urine densities of all groups (mean ± SD).

Groups	Group C	Group S	Group A	Group UPO	Group UPT
$Density_{0th\ week}$	1019.50 ± 3.27	1035.33 ± 12.75	–	–	–
$Density_{2th\ week}$	1020.10 ± 2.73	1034.50 ± 12.24	1020.60 ± 13.90	1027.60 ± 42.86	1027.50 ± 5.14
$Density_{4th\ week}$	–	–	1013.33 ± 4.97	1027.00 ± 5.14[a]	1012.40 ± 3.64

C: control; S: sham; A: alfuzosin; UPO: ureteropelvic junction obstruction; UPT: ureteropelvic junction obstruction + alfuzosin treatment.
[a]$p < 0.05$ Group UPO versus others.

There were not any difference ($p > 0.05$) between groups (except UPO) regarding urine β_2 microglobulin levels, but Group UPO had higher microglobulin levels than the rest of the groups ($p = 0.041$). Only Group UPO's before and after experiment values were significantly higher in repeating measurements within-group ($p = 0.041$). However, there is no statistically significant difference between the before and after values of Group UPT ($p > 0.05$) (Table 2).

While there is no notable difference ($p > 0.05$) regarding urine electrolytes or density (excluding the comparison between Group UPO and Group S about their repeating Na, P, and K value measurements), it was observed that urine density in Group UPO has increased significantly ($p = 0.048$) with regards to the repeating measurements within-groups from before and after the experiment and that other groups did not undergo any change regarding their start and end values (Table 3).

After evaluating the blood electrolyte levels of the groups, it was found that K, P, Cl, Ca, BUN, and creatinine levels in group UPO were in normal and statistically significant levels ($p = 0.01$) compared to those of Group UPT administered with alfuzosin (Table 4).

Analyzing the serum and urine TGF-β_1 values, the lowest values for serum was in Group A (45.10 ± 58.83) and for urine in Group S (45.10 ± 58.83); while the highest values for both parameters were in Group UPO. While there is no statistical difference ($p = 0.494$) in serum values between the group with stenosis (UPO) and the one who got treatment after stenosis (UPT), it was significantly lower (86.96 ± 69.23 and 112.28 ± 58.65). The urine TGF-β_1 value of Group UPT were again significantly lower than the value of UPO ($p = 0.012$) (Table 5).

No significant difference was identified in ARI values of Group A ($p > 0.05$). With Group UPT, however, ARI values measured after stenosis (ARI_2) were statistically significant higher than the control ARI values ($p = 0.001$). The ARI values measured after alfuzosin treatment in the same group (ARI_4) were statistically and significantly lower ($p = 0.046$) than the values measured after the inducement of UP junction stenosis (ARI_2) (Figure 2, Table 6).

AP diameter measurements of Groups UPT and UPO were statistically higher than the other groups' repeating values ($p = 0.001$) and even though there was not a statistically significant difference between the measurements of aforementioned groups, AP diameter of Group UPT was lower (15.84 ± 0.58 and 12.16 ± 0.39). Moreover, the starting values of both Groups UPO and UPT and the AP diameter were lower than the subsequent measurements ($p = 0.001$). Group UPT's AP diameter value decreased statistically and significantly ($p = 0.001$) after treatment (from 14.94 ± 1.21 to 12.16 ± 0.39) (Table 6).

Table 2. Weekly urine β_2 microglobulin levels (mg/dl) of all groups (mean ± SD).

$Group/_{week}$	β_2 $Microglobulin_0$	β_2 $Microglobulin_2$	β_2 $Microglobulin_4$
Group C	0.38 ± 0.12	0.41 ± 0.21	–
Group S	0.37 ± 0.15	0.38 ± 0.12	–
Group A	–	0.37 ± 0.15	0.38 ± 0.11
Group UPO	–	1.23 ± 1.59	1.99 ± 2.59[a]
Group UPT	–	0.31 ± 0.08	0.38 ± 0.14

C: control; S: sham; A: alfuzosin; UPO: ureteropelvic junction obstruction; UPT: ureteropelvic junction obstruction + alfuzosin treatment.
[a]$p < 0.05$ for Group UPO.

While there was no difference between groups and the repeating resistive indexes of the groups ($p > 0.05$), it has contributed to identifying renal artery resistive index values in rats as 0.43–0.45 on average (Table 6).

When the groups are examined histopathologically, it is seen that there is no difference regarding any of the histopathological criteria analyzed between Groups C, S, and A. No statistically significant difference was identified between groups with regards to proximal and distal tubule necrosis and interstitial edema ($p > 0.05$). The values for glomerulus sclerosis and collapse, proximal and distal tubule dilation, epithelial degeneration, interstitial fibrosis and inflammation, and vascular congestion for Group UPO were significantly and statistically higher than the rest of the groups ($p = 0.001$). While the values of Group UPT for these parameters are higher than the ones of Groups S, C, and A ($p = 0.03$, 0.02 and 0.02, respectively), there was a declining difference both in number and in the degree of the damage compared to Group UPO (Figure 3) (p values = between 0.003 and 0.020). But, no statistical difference was identified for glomerulus collapse, distal tubule dilation, and interstitial fibrosis (Table 7).

Discussion

UPO is the obstruction of urine passage from pelvis to ureter due to anatomic or functional reasons and in

Table 3. Weekly urine electrolyte levels of all groups (mean ± SD).

Electrolyte/group	Group C	Group S	Group A	Group UPO[a]	Group UPT
Na_0 (mmol/L)	18.9 ± 7.87	24.41 ± 10.66	–	–	–
Na_2 (mmol/L)	18.71 ± 7.68	24.48 ± 10.85	5.6 ± 3.2	14.00 ± 1.58[a]	18.60 ± 4.03
Na_4 (mmol/L)	–	–	5.60 ± 3.2	10.6 ± 3.64	6.40 ± 3.28
K_0 (mmol/L)	77.83 ± 19.13	153.48 ± 48.95	–	–	–
K_2 (mmol/L)	77.63 ± 18.71	152.11 ± 49.07	87.29 ± 48.48	70.73 ± 26.31	142.62 ± 71.79
K_4 (mmol/L)	–	–	87.29 ± 48.48	118.25 ± 46.40[a]	56.48 ± 16.73
Cl_0 (mmol/L)	27.65 ± 11.16	54.36 ± 41.20	–	–	–
Cl_2 (mmol/L)	24.15 ± 14.84	54.13 ± 41.16	14.60 ± 3.97	17.40 ± 3.97	30.80 ± 7.08
Cl_4 (mmol/L)	–	–	14.60 ± 3.71	22.80 ± 7.98	13.60 ± 7.02
P_0 (mg/dl)	80.37 ± 21.64	155.26 ± 91.59	–	–	–
P_2 (mg/dl)	80.12 ± 20.80	152.38 ± 87	36.17 ± 58.73	45.37 ± 11.22	31.89 ± 16.63
P_4 (mg/dl)	–	–	36.17 ± 58.73	42.53 ± 21.71[a]	18.95 ± 7.65
Ca_0 (mg/dl)	5.62 ± 2.15	7.52 ± 4.05	–	–	–
Ca_2 (mg/dl)	5.68 ± 2.05	7.44 ± 3.75	4.32 ± 3.03	3.55 ± 1.72	4.05 ± 2.34
Ca_4 (mg/dl)	–	–	4.32 ± 3.03	3.77 ± 1.21	3.76 ± 1.79

C: control; S: sham; A: alfuzosin; UPO: ureteropelvic junction obstruction; UPT: ureteropelvic junction obstruction + alfuzosin treatment.
[a]$p < 0.05$ Group UPO versus others.

Table 4. Blood electrolyte levels of all groups (mean ± SD).

Electrolyte/group	Group C	Group S	Group A	Group UPO	Group UPT[a]
Na (mmol/L)	142.40 ± 17.87	143.20 ± 1.64	144.50 ± 1.64	143.80 ± 2.94	143.60 ± 0.89
K (mmol/L)	5.50 ± 0.41	5.52 ± 0.46	5.04 ± 0.34	7.51 ± 0.52	5.45 ± 0.38
Cl (mmol/L)	104.02 ± 1.71	104.12 ± 1.73	102.15 ± 1.88	96.68 ± 1.08	101.62 ± 1.19
P (mg/dl)	6.33 ± 0.77	6.28 ± 0.76	7.23 ± 1.41	3.90 ± 0.75	6.33 ± 1.04
Ca (mg/dl)	9.40 ± 0.27	9.43 ± 0.24	9.10 ± 0.16	10.20 ± 0.38	9.44 ± 0.39
BUN (mg/dl)	32.02 ± 0.70	32.00 ± 0.65	30.13 ± 1.84	80.76 ± 15.92	36.62 ± 5.37
Creatinine (mg/dl)	0.32 ± 0.06	0.31 ± 0.16	1.84 ± 3.60	1.01 ± 0.18	0.44 ± 0.03

C: control; S: sham; A: alfuzosin; UPO: ureteropelvic junction obstruction; UPT: ureteropelvic junction obstruction + alfuzosin treatment.
[a]$p < 0.05$ Group UPT versus Group UPO.

Table 5. TGF-β_1 values of all groups (expression unit) (mean ± SD).

TGF-β_1 level	Group K	Group S	Group A	Group UPO	Group UPT
Serum	108.33 ± 56.44	108.15 ± 56.29	45.10 ± 58.83	112.28 ± 58.65	86.96 ± 69.23[a]
Urine	57.80 ± 14.24	45.10 ± 58.83	88.81 ± 55.66	119.14 ± 28.58	56.46 ± 44.11[b]

C: control; S: sham; A: alfuzosin; UPO: ureteropelvic junction obstruction; UPT: ureteropelvic junction obstruction + alfuzosin treatment.
[a]Group UPT serum TGF-β_1 levels lower than Group UPO but $p = 0.494$.
[b]$p < 0.05$ Group UPT versus UPO.

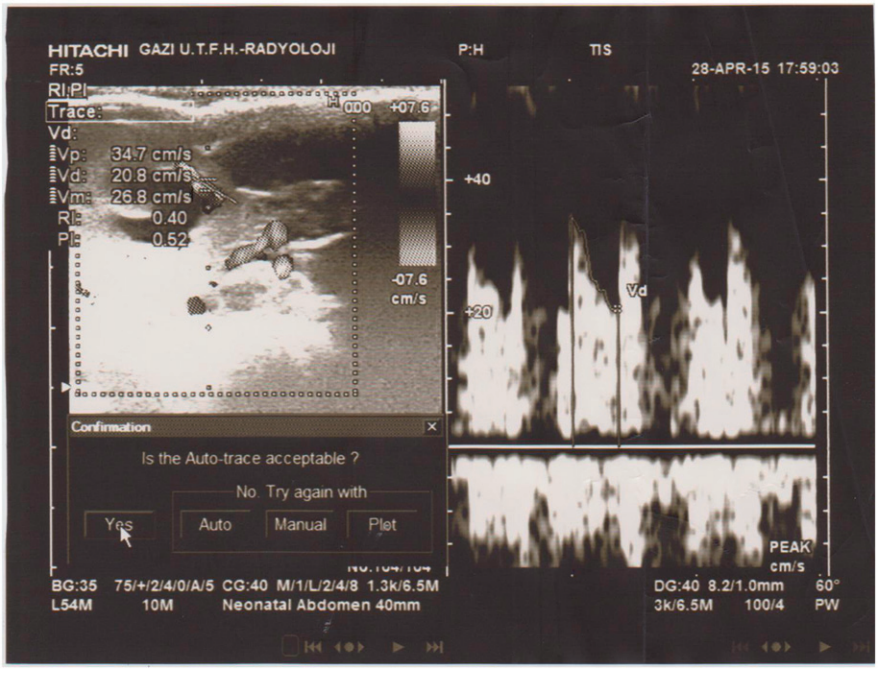


Figure 2. Increased anteroposterior diameter and calculated ARI values (0.52) in a UPT group rat by US.

Table 6. Weekly radiological evaluation for anteroposterior (AP) diameter (mm) of all groups (mean ± SD).

USG/groups	Group C	Group S	Group A	Group UPO	Group UPT
AP_0	9.96 ± 0.58	–	–	–	–
AP_2	–	9.96 ± 0.58	10.76 ± 1.92	14.90 ± 1.05	14.94 ± 1.21
AP_4	–	9.90 ± 0.55	8.91 ± 1.45	15.84 ± 0.58[a]	12.16 ± 0.39[ab]
ARI_0	0.41 ± 0.02	–	–	–	–
ARI_2	–	0.45 ± 0.09	0.40 ± 0.09	0.44 ± 0.08	0.46 ± 0.01
ARI_4	–	0.43 ± 0.36	0.41 ± 0.09	0.46 ± 0.03	0.39 ± 0.03

C: control; S: sham; A: alfuzosin; UPO: ureteropelvic junction obstruction; UPT: ureteropelvic junction obstruction + alfuzosin treatment.
[a]$p < 0.05$ Groups UPO and UPT versus others.
[b]$p < 0.05$ Group UPT versus Group UPO.

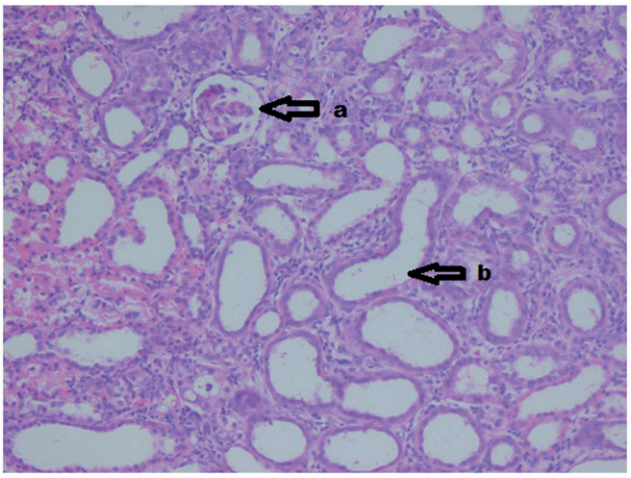


Figure 3. Glomerular collapse (a) and tubular dilatation (b) were shown in UPT group (HE 100×).

Table 7. Histopathological evaluation of all groups.

Groups	Group C	Group S	Group A	Group UPO[a]	Group UPT[b]
Glomerulus sclerosis	0	0	0	5	0
Glomerulus collapse	0	0	0	5	2
Proximal tubule necrosis	0	0	0	0	0
Proximal tubule dilation	0	0	0	6	3
Proximal tubule Epithelial degeneration	0	0	0	6	4
Distal tubule necrosis	0	0	0	0	0
Distal tubule dilation	0	0	0	6	3
Distal tubule Epithelial degeneration	0	0	0	6	3
Interstitial edema	0	0	0	0	0
Interstitial fibrosis	0	0	0	3	1
Interstitial inflammation	0	0	0	4	2
Vascular congestion	0	0	0	5	1

C: control; S: sham; A: alfuzosin; UPO: ureteropelvic junction obstruction; UPT: ureteropelvic junction obstruction + alfuzosin treatment.
[a]$p < 0.05$ Group UPO versus others.
[b]$p < 0.05$ Group UPT versus Group UPO.

various degrees. It is one of the common reasons for obstructive nephropathy. Failure to transfer the peristaltic waves down due to the abnormal muscular activity in the junction is thought to be the main cause for the intrinsic type which is also the most common.[16] The study conducted by Portincasa et al. on this subject has shown that there is a significant deterioration in the smooth muscle contraction at the ureteropelvic junction in both obstructive and functional UPO cases.[17]

USG and scintigraphy are major screening methods in diagnosing UPO, evaluating the patients after surgery, and monitoring patients who have not undergone a surgical operation yet.[18,19] Biochemical indicators such as TGF-β_1 and β_2 microglobulin, urine, and blood electrolytes are also utilized in diagnosing and monitoring UPO in the recent years.[20–25]

Alfuzosin, one of the selective α_1 blockers, is a pharmacological agent used in prostate hypertrophy. Thanks to its ability to inhibit smooth muscle contractions, it has been utilized recently for ensuring smooth passage of ureteral calculi.[12,13] Our aim was to investigate whether alfuzosin has protective effects on tubules and kidneys in partial UPO model in order to benefit from its ability to reduces intraluminal pressure in the ureter by selectively blocking α_{1a} especially in the proximal ureter.

Renal tubular damage occurs in UPO obstruction cases.[2] It was shown that the number of aquaporins which ensures that the urine is concentrated in collector channels with proximal and distal tubules, sodium, and urea transporter proteins decrease as a symptom of renal tubular function disorder. Renin angiotensin-aldosterone (RAA) system causes decrease in the number of AQP and sodium transporter proteins in UPO and deterioration in the urine concentration ability.[3–6] Indeed, it was seen that the urine density measured after the alfuzosin therapy in Group UPT were statistically and significantly lower than both their first values and the ones of Group UPO ($p < 0.05$). No notable statistical difference ($p > 0.05$) regarding urine electrolytes in any group (excluding the comparison between Group UPO and Group S about their repeating Na, P, and K value measurements).

β_2 Microglobulin is a substance partially reabsorbed when it reaches the tubules after being filtered by the glomeruli. In tubular renal diseases, β_2 microglobulin filtered in glomeruli cannot be reabsorbed from the tubules, and thus its level in urine augments. In the studies conducted by Bartoli et al. with children with UPO, they have showed that β_2 microglobulin levels are high in functional and obstructive congestion and that it drops in cases where the stenosis is eliminated with surgery.[26] In this experimental model, the increase of the urine values in Group UPO after stenosis

demonstrates: β_2 microglobulin levels decreased back to its normal limits with alfuzosin treatment in Group UPT.

Serum K, P, Cl, Ca, BUN, and creatinine levels in Group UPT administered with alfuzosin reaches back to its statistically significant normal values compared to Group UPO, and that its renal functions were conserved with alfuzosin treatment.

TGF-β_1 is one of the main mediators of tubulointerstitial fibrosis in UPO.[27] TGF-β1 may be used in diagnosing and monitoring UPO as a noninvasive and simple method since renal damage also increases. High pressure in upper urinary tract in hydronephrosis and obstruction cases causes various molecular and histopathological changes and TGF-β1 increase by stimulating the RAA system and inflammation.[28,29] TGF-β1 isolated in serum in UPO cases were never before utilized, examined in this study for the first time, and detected in high levels in study groups. In this study, urine TGF-β_1 levels of both groups with UPO were seen to be statistically higher compared to the other groups ($p < 0.05$) while the kidneys were under hydronephrotic monitoring both USG and macroscopically during the exploration made while terminating the experiment. The fact that both urine and blood TGF-β_1 values were decreased after treatment is interpreted as alfuzosin reduces tubulointerstitial fibrosis. While Merriki and Sager's studies found a higher urinary TGF-β_1 in children with UPO, these levels dropped after surgical treatment or spontaneous recovery.[28,29]

AP diameters and renal ARI measurements were also utilized in our study in order to support the diagnostic accuracy of USG. The aim of this study is to constitute a reference for future studies by determining the average ARI values of standard rats and the ones induced with UPO in medical literature ($ARI_0 = 0.49 \pm 0.02$).

AP diameter values turned out to be more significant in diagnosing UPO and monitoring its progression compared to ARI. Highest AP values were found in Groups UPT and UPO. These high AP diameter values and urine collection for 24 h serve as a proof that the UPO model is not a fully obstructive model. It was also shown that the alfuzosin treatment administered after the stenosis in Group UPT prevents or reverses the augmentation of AP diameter.

All ARI values were similar in all groups at the start in this study. However, this value has risen following the UPO and it was reduced with selective α_{1a} blocker alfuzosin treatment in Group UPT. Even though the average ARI values were similar, this decrease due to the alfuzosin treatment gives rise to the thought that it might be because of an inhibition ensured sympathicomimetically by alfuzosin indirectly, if not directly.[12,13] The study made by Karakuş et al., it was shown that ARI is fit to be utilized in diagnosing UPO and that enalapril, an ACE inhibitor used on the treatment group, maintains renal blood flow with its effect on the RAA system and decreases ARI values.[30]

Glomerulus sclerosis and collapse, proximal and distal tubule dilation, epithelial degeneration, interstitial fibrosis and inflammation, and vascular congestion were reduced statistically in Group UPT by histopathologically ($p < 0.05$). This situation leads us to believe that alfuzosin has protective effect on kidneys.

Villa et al. study was shown α_1 blockers decreases the intraluminal ureter pressure in partial ureteral obstruction model where *in vivo* intraluminal ureteral muscular waves were being measured.[11] Kobayashi et al. was found rat ureter contractions induced with α-adrenoreceptors were suppressed by α_{1a} adrenoreceptor antagonists.[31] It was revealed that 10 mg/kg alfuzosin treatment of rats with erectile dysfunction and partial bladder outlet obstruction has antioxidant effect, restored NO release, and decreased sympathomimetic effecs.[12] Besides its aforementioned effect of reducing the pressure on the urinary system, it is thought that alfuzosin provides protective benefits on kidneys and the RAA system with its antioxidant efficiency and by decreasing the sympathicomimetic tonus. The limitations of this study are made without measurement antioxidant parameters, *in vivo* intraluminal pressure study. These parameters could be measured in other studies.

This study shows the protective effects of alfuzosin treatment on kidneys in rats induced with moderate partial chronic UPO by reducing the pressure on especially the urinary system and by decreasing both β_2 microglobulin and TGF-β_1 levels histopathologically.

Disclosure statement

The authors report no conflicts of interest.

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