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The effect of nanomicellar curcuminoids on renal ischemia/reperfusion injury and the expressions of COX-2 and Na^+/K^+-ATPase in rat's kidney

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ABSTRACT

Introduction: Renal Ischemia/Reperfusion (I/R) causes acute kidney injury known by impaired renal function, which has partially been connected to kidney apoptosis as well as the impairment of Cyclooxygenase-2 (COX-2) and Na^+/K^+-ATPase signaling. Curcuminoids have been proposed to have potential renoprotective effects. Thus, the present research work aimed to assess the effect of Nanomicellar Curcuminoids (NC) in a rat model of renal I/R.

Methods: Adult male Sprague-Dawley rats were allocated to three treatment groups (n=5/group). NC at the dose of 25 mg/kg/i.p or its vehicle was administered 60 min before renal ischemia induction. Then, the animals were subjected to bilateral renal ischemia for 60 min and reperfusion for 24 h. Subsequently, blood samples were collected to assess Blood Urea Nitrogen (BUN) and Creatinine (Cr) levels. In addition, kidneys were isolated to evaluate renal histopathology, caspase-3 cleavage, and COX-2 and Na^+/K^+ -ATPase pump levels.

Results: The results showed that NC improved kidney function ($P<0.0001$) and attenuated I/R-induced histopathological injuries ($P<0.0001$) and caspase-3 cleavage ($P<0.01$). However, the downregulation of renal COX-2 and Na^+/K^+ -ATPase expression induced by I/R was not restored by the renoprotective dose of NC.

Conclusion: The findings of the present study indicated that the renoprotective effect of NC in the renal I/R rat model coincided with the inhibition of histopathological injuries and apoptosis, but not with compensation for renal COX-2 and Na^+/K^+ -ATPase downregulation.

Keywords:
Curcuminoids
Nanoparticle
Renal ischemia/reperfusion
Na^+/K^+ -ATPase
COX-2
Rat

Introduction

Ischemia followed by reperfusion (I/R) leads to serious organ damage (Soares et al., 2019). Renal I/R is a common outcome of clinical procedures such as renal transplantation and partial nephrectomy (Lugo-Baruqui et al., 2019; Soares et al., 2019). Furthermore, renal I/R is an important cause of acute renal failure characterized by low rates of glomerular filtration and tubular necrosis (Shiva et al., 2020).

During ischemia, mitochondrial oxidative phosphor-

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ylation is inhibited due to the lack of oxygen. This phenomenon impairs ATP synthesis and reduces the activity of energy-dependent pumps, which can lead to cell death (Malek and Nematbakhsh, 2015). ATP depletion may also affect Na^+/K^+-ATPase as the main pump involved in renal sodium reabsorption (Féraille and Doucet, 2001; Matsuzaki et al., 2007; Sampaio et al., 2018). Moreover, renal I/R activates the inflammatory cascades that contribute to renal injury. Therefore, modulation of the inflammatory process is considered a therapeutic approach in renal I/R (Malek and Nematbakhsh 2015). Cyclooxygenase-2 (COX-2) is considered a pro-inflammatory factor involved in renal I/R injury (Nørregaard et al., 2015). Thus, the use of COX-2 inhibitors has been suggested to benefit renal I/R (Goetz Moro et al., 2017). However, some studies have revealed a decrease in the expression of COX-2 during renal I/R (Nørregaard et al., 2015). Hence, it is unknown whether the decrease in the COX-2 level is a protective or damaging factor (Nørregaard et al., 2015).

Recently, scientists have been inspired to evaluate the effects of herbal agents on I/R. Turmeric has been extensively used as a traditional medicine to treat various disorders such as biliary disorders, liver diseases, anorexia, common cold, diabetic wounds, and rheumatism (Amalraj et al., 2016). The active ingredients of turmeric called curcuminoids include curcumin, Demethoxycurcumin (DMC), and Bisdemethoxycurcumin (BDMC). Curcuminoids have been suggested to exert several protections such as renoprotective effects (Amalraj et al., 2016). Howbeit, the protective effects of curcuminoids are limited by their low bioavailability and low aqueous solubility (Amalraj et al., 2016; Liu et al., 2016b). Therefore, several efforts have been made to improve the bioavailability of curcuminoids to enhance their therapeutic effects (Amalraj et al., 2016; Dawidczyk et al., 2014). Nano-micelles have been considered effective tools to encapsulate drugs with low water solubility (Williams et al., 2013). The micelle core-shell structure inhibits water penetration, thereby providing a suitable environment for curcumin compared to its native form (Hatamipour et al., 2019). A new formulation of Nanomicellar Curcuminoids (NC) with enhanced solubility, oral bioavailability, and stability of all the three major curcuminoids; i.e. curcumin, DMC, and BDMC, has been fabricated (Hatamipour et al., 2019) and commercialized with the trade name of SinaCurcumin by Exir Nano Sina Company (Tehran, Iran), which is currently utilized as a curcuminoid supplement. Several reports have indicated the beneficial effects of this compound in multiple clinical trials (Prasad et al., 2014; Wright et al., 1982).

Considering the advantages of the nanomicelles of curcuminoids, the present study aims to investigate the renoprotective effects of SinaCurcumin® as an NC drug on kidney function, histopathological injuries, renal caspase-3 cleavage, and COX-2 and Na^+/K^+-ATPase levels in a rat model of renal I/R.

Materials and Methods

Materials

NC registered as SinaCurcumin® was purchased from Exir Nano Sina Company, Tehran, Iran (IRC: 1228225765). The characterization of SinaCurcumin® has been published previously (Hatamipour et al., 2019). The western blot analysis antibodies, Na^+/K^+-ATPase, GAPDH, and anti-goat IgG Horseradish peroxidase-conjugated antibodies were prepared by the Santa Cruz Company. Caspase-3, COX-2, and secondary Anti-Rabbit IgG HRP-conjugated antibodies were obtained from Cell Signaling Technology (Danvers, MA, USA). Besides, Polyvinylidene Difluoride (PVDF) membrane was purchased from Millipore and the protease inhibitor was supplied by Pierce. Additionally, Amersham Enhanced Chemiluminescence (ECL) select reagent kit was obtained from GE Healthcare Life Sciences, UK. Other reagents were obtained from usual commercial sources.

Experimental animals

Adult male Sprague-Dawley rats (weight 220-250 g) were purchased from the Laboratory Animal Center of Shiraz University of Medical Sciences, Shiraz, Iran. The rats had access to water and food *ad libitum* and were housed under controlled temperature (20±2°C) and lighting (07:00 to 19:00 h). Ethical approval was obtained from the Local Committee for the Ethics of Scientific Research of Shiraz University of Medical Sciences (approval No. IR.SUMS.REC.1399.556). All animal experiments were carried out in accordance with the National Institutes of Health guide for the care and use of laboratory animals (NIH publications No. 8023, revised 1978).

The animals were randomly allocated into three treat-

ment groups (n=5/group):

1. Sham group: received saline as the vehicle of NC one h before laparotomy without ischemia induction.

2. I/R group: received saline as the vehicle of NC one h before bilateral I/R induction.

3. NC + I/R: received NC 25 mg/kg/ip one hour before I/R induction.

Renal ischemic–reperfusion induction

Renal I/R was induced as described previously (Gholampour et al., 2019; Karimi et al., 2021). Briefly, the animals were anesthetized using ketamine (50 mg/kg/i.p.) and xylazine (10 mg/kg/i.p.). Following a middle laparotomy, the rats were subjected to bilateral renal pedicle occlusion for 60 min using a microaneurysm vascular clamp. After 60 min, the clamps were removed and the wounds were sutured in two layers. The animals experienced a 24 h reperfusion afterward. The animals in the sham group underwent laparotomy without pedicel clamping.

Assessment of the plasma levels of blood urea nitrogen and creatinine

Following 24 h of reperfusion, the animals were anesthetized using ketamine (50 mg/kg/i.p.) and xylazine (10 mg/kg/i.p.). Then, blood samples were obtained from their tails for biochemical assessments. The blood samples were centrifuged at 4000 rpm for 15 min, and the plasma was stored at -20°C for the assessment of Blood Urea Nitrogen (BUN) and Creatinine (Cr) levels. The plasma levels of BUN and Cr were analyzed by an auto-analyzer (RA-1000 Technicon, America).

Histological analysis and injury scoring

Following 24 h of reperfusion, the right kidney was isolated in anesthetized rats to assess the histological parameters in all the study groups (n=4 randomly assigned). The isolated kidney was post-fixed in formalin 10%, dehydrated with alcohol, and embedded in paraffin for Hematoxylin-Eosin (H &E) staining. The tissue block was then sliced into 4 µm sections and stained with H&E and the histopathological injury scoring was performed using light microscopy. The percentage of injury was calculated using several parameters including the shedding of brush border, tubular necrosis, vascular congestion, exfoliation of epithelial cells, and cast deposition. The degree of renal damage was determined using 10 randomly selected fields in both the cortex and medullary area for each animal using the following criteria: 0=no damage, 1=minor (<20% abnormality of the cortex or outer medulla), 2=moderate (21-40% abnormality of the cortex or outer medulla), 3=severe (41-60% injury of the cortex or outer medulla), and 4=more severe defects (>81% involvement of the cortex or outer medulla).

Western blot analysis

Following 24 h of reperfusion, the left kidney was isolated in anesthetized rats to assess caspase-3, COX-2, and Na^+/K^+-ATPase levels (n=3 randomly assigned). The kidney tissues were homogenized and lysed in cold RIPA (Radioimmunoprecipitation assay) lysis buffer containing protease and phosphatase inhibitors. The lysates were then centrifuged at 12000 rpm at 4°C for 25 min. Afterwards, the supernatant was isolated and its protein content was determined using the Lowry method. The Western blot technique was performed as previously described (Amiri et al., 2016; Moosavi et al., 2014). Briefly, equal protein samples (40 µg/sample) were electrophoresed on a 10% Sodium Dodecyl Sulfate PolyAcrylamide Gel Electrophoresis (SDS–PAGE) and were transferred onto PVDF membranes (Millipore, Burlington, MA, United States) using the Bio-Rad transfer system (Bio-Rad, USA). The blots were blocked with 5% Bovine Serum Albumin (BSA) in TBST (100 mM Tris, 2.0% NaCl pH 7.6, 1% Tween-20) at room temperature for one h. The membranes were then exposed to primary antibodies including caspase-3 (1/1000), COX-2 (1/2000), Na^+/K^+-ATPase (1/2000), and GAPDH (1/2000) overnight at 4°C. After that, the membranes were washed three times with TBST (10 min each) and were exposed to their corresponding HRP-conjugated secondary antibodies at room temperature for one h. After washing with TBST, the bands were revealed using enhanced chemiluminescence (ECL select; GE Healthcare) and photographic films in a dark room. Image J software from NIH (Bethesda, MD, USA) was used to quantify the intensities of the protein bands.

Statistical analysis

The data have been presented as mean ± Standard Error of Measurement (SEM), $P<0.05$ was defined statistically significant. Data analysis was performed using GraphPad Prism version 6 (GraphPad software, San

Diego, CA). The data were analyzed by one-way ANOVA followed by Tukey's test for multiple comparisons.

Results

NC treatment improved renal function

Renal I/R led to a significant increase in the plasma levels of BUN (F (2, 12) = 227; $P<0.0001$) and Cr (F (2, 12) = 172; $P<0.0001$), indicating renal function disruption. Treatment with NC 25 mg/kg/ip 60 min before ischemia induction significantly decreased the plasma

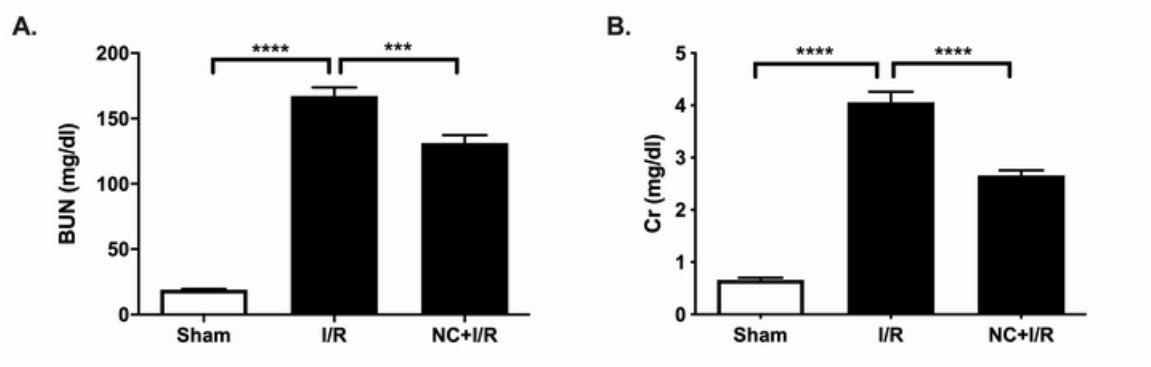

FIGURE 1. The plasma levels of BUN (A) and Cr (B) in the sham, I/R, and NC 25 mg/kg + I/R groups. The data have been presented as mean ± SEM. $^{***}p<0.001$ and $^{****}p<0.0001$ represent statistical differences between the study groups.

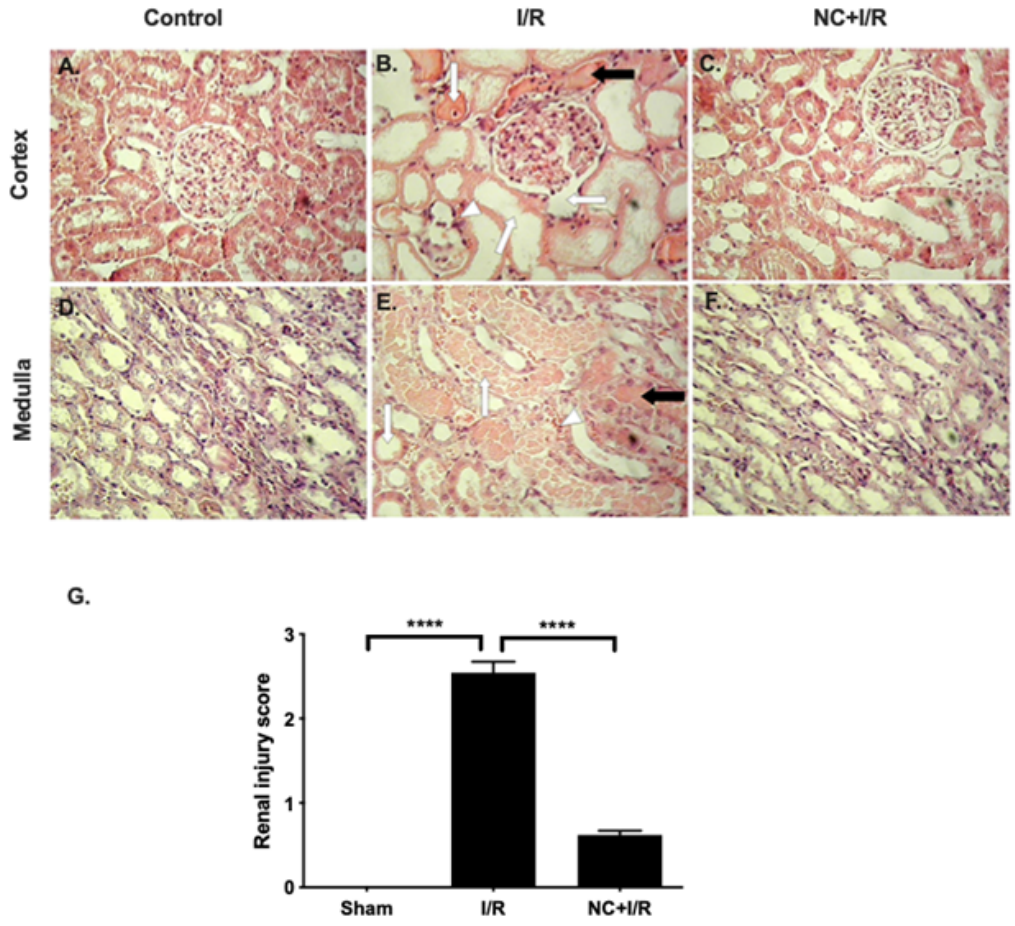

FIGURE 2. The histopathological results following renal I/R. Light microscopic images of renal cortex and medulla stained with H&E (400X magnification) in the sham (A and D), I/R (B and E), and NC+I/R (C and F) groups. In the I/R group, the enlargement of the bowman space has been shown with a white horizontal arrow (B), destruction of the proximal tubule with a white upward arrow (B and E), damage to the thick ascending limb with a white downward arrow (C), intratubular cast formation with a black arrow (B and E), and vascular congestion with a triangle arrow (B and E). The quantitative injury score of H&E staining has been presented in Fig. 3G. The data have been presented as mean ± SEM. $^{****}p<0.0001$ represents statistical differences between the study groups.

levels of BUN (P<0.001, Figure 1A) and Cr (P<0.0001, Figure 1B) compared to the I/R animals.

NC administration reduced the histological lesions

The representative histopathological images of the cortical and medullary kidneys of the sham, I/R, and I/R + NC groups have been depicted in Figure 2 A-F. As shown in Figure 2B and 2E, renal I/R led to tubular and vascular enlargement (including the bowman space), destruction of proximal and thick ascending tubules, intratubular cast formation, and vascular congestion in the cortex and medulla (Figure 2B and 2E). These histopathological changes were attenuated following the treatment with NC 25 mg/kg (Figure 2C and 2F). The total histopathological injury scores have been presented in Figure 2G. The results of one-way ANOVA revealed a significant difference among the study groups (F (2, 9) = 274; P<0.0001). As shown in Figure 2G, NC administration significantly improved renal injury compared to the I/R group (P<0.0001).

The protein levels of cleaved caspase 3, Cox-2, and Na^+/K^+-ATPase following NC administration

The amount of cleaved caspase-3 protein was determined using the western blot technique, and the results have been depicted in Figure 3A. The quantitative analysis of cleaved caspase-3 has also been shown in Figure 3B. The results of one-way ANOVA indicated a significant difference among the study groups (F (2, 6) = 6.75, p=0.0291). Additionally, the results of post-hoc Tukey's test showed a significant difference between the sham and renal I/R groups (P<0.05), while there was no significant difference between the sham and NC + I/R groups. These results revealed the anti-apoptotic effect of NC in the rat model of renal I/R.

The level of COX-2 protein was determined using the western blot technique, and the results have been presented in Figure 3A. Besides, the quantitative ratios of COX-2 in the sham, I/R, and NC + I/R groups have been shown in Figure 3C. The results of one-way ANOVA indicated a significant difference among the study groups (F (2, 6) = 13.9, p=0.0056). The results of post-hoc Tukey's test also demonstrated a significant difference between the sham and renal I/R groups (P<0.01). However, NC treatment was not able to restore the renal COX-2 downregulation.

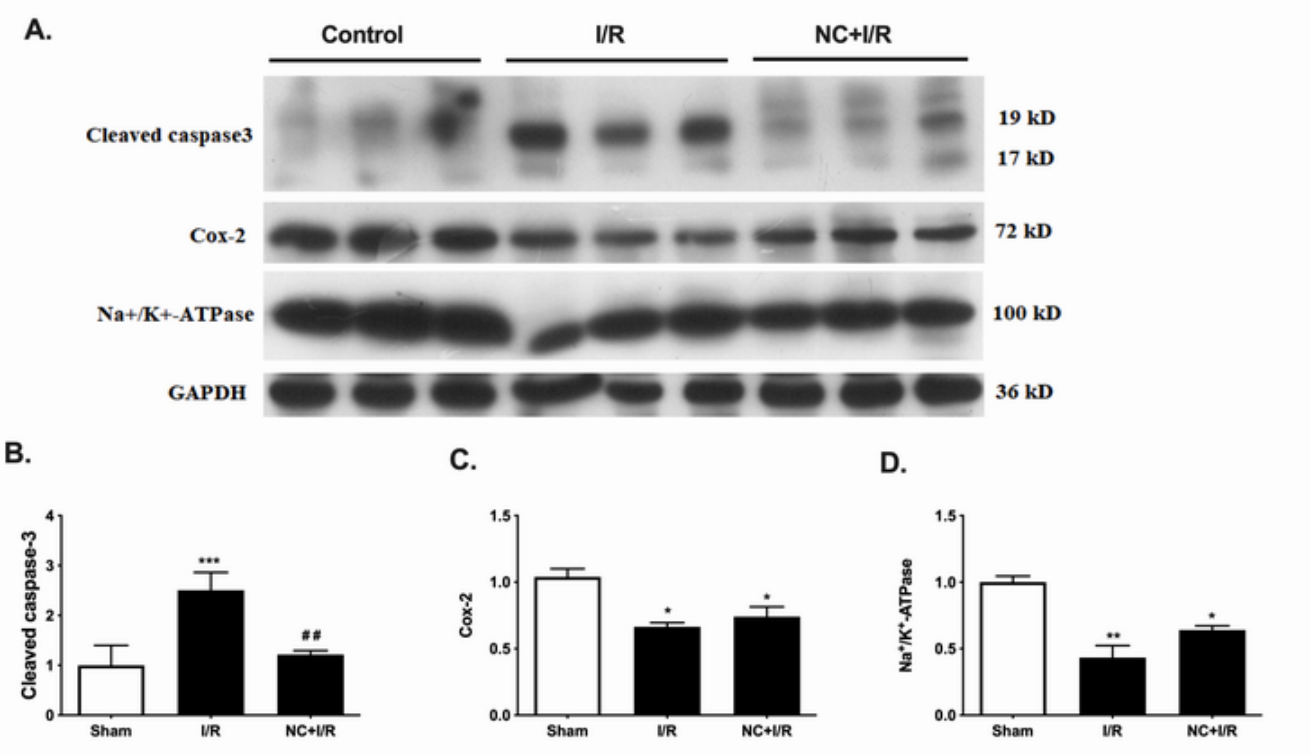

FIGURE 3. The protein levels of renal cleaved caspase-3, Cox-2, Na^+/K^+-ATPase, and GAPDH in the sham, I/R, and NC+I/R groups (A). The quantitative ratio of cleaved caspase-3 (B), Cox-2 (C), and Na^+/K^+-ATPase (D) in different groups. Data have been shown as mean ± SEM. $^{*}P$<0.05, $^{**}P$<0.01, and $^{***}P$<0.001 represent statistical differences between the control and other groups. $^{\#\#}P$<0.01 represents differences between the I/R and NC + I/R treated groups.

The immunoblot results of renal Na^+/K^+-ATPase have been shown in Figure 3A. In addition, the quantitative ratios of Na^+/K^+-ATPase in different groups have been presented in Figure 3D. The results of one-way ANOVA revealed a significant difference among the study groups ($F(2, 6) = 22.4$, $p=0.0016$). The results of post-hoc Tukey's test also revealed that renal I/R significantly reduced Na^+/K^+-ATPase expression in the kidney ($P<0.01$) and the renoprotective dose of NC did not prevent I/R-induced Na^+/K^+-ATPase downregulation.

Discussion

The present study results revealed an increase in the plasma levels of BUN and Cr following renal I/R. However, a single CN administration at the dose of 25 mg/kg/ip significantly decreased the levels of these parameters, representing the beneficial effect of NC on renal function. Previous studies have mostly dealt with the renoprotective effect of curcumin (Kaur et al., 2016; Liu et al., 2017; Liu et al., 2016a; Najafi et al., 2015; Zhang et al., 2018). Nonetheless, curcumin is not the only bioactive curcuminoid. Moreover, the previously reported protective doses of curcumin were quite higher than that administered in the present investigation. For instance, in the study performed by Najafi et al., rats were administered with a 20 mg/kg curcumin intraperitoneal injection following ischemia induction. Following a 24 h reperfusion, curcumin could not significantly decrease the serum level of Cr in I/R animals (Najafi et al., 2015). Other studies reported that curcumin at the dose of 100 (Kar et al., 2020) and 200 mg/kg/i.p. (Aydin et al., 2014; Karahan et al., 2016) might protect rats against renal I/R injury (Aydin et al., 2014; Karahan et al., 2016). Therefore, the triple curcuminoid formulation and the higher bioavailability of NC used in the present study (Hatamipour et al., 2019) might potentiate the protective effects and lower the efficient dose. Furthermore, the histopathological results showed an improvement in tubular necrosis, exfoliation of the epithelial cells of the proximal tubule and the thick ascending limb, cast formation, and vascular congestion following a single NC treatment.

The results of the current research disclosed that renal I/R led to the downregulation of the Na^+/K^+-ATPase pump. Generally, Na^+/K^+-ATPase is highly expressed in the kidney, because the maintenance of the Na^+ gradient is fundamental to reabsorb amino acids and glucose as well as to regulate blood pH and electrolytes (Clausen et al., 2017). Additionally, the energy metabolic dysfunction induced by ischemia seems to be closely connected to the Na^+/K^+-ATPase action (Matsuzaki et al., 2007; Sampaio et al., 2018). However, the nephroprotective effect of NC did not coincide with the restoration of Na^+/K^+-ATPase pump expression in the present study. Therefore, it seems that the restoration of the Na^+/K^+-ATPase pump is not involved in the renoprotective effect of NC.

Previously, a strong association was recommended between COX-2 and renal I/R injury (Nørregaard et al., 2015). Therefore, the COX-2 level was assessed within the renal tissue in the present research. The results revealed that renal I/R downregulated COX-2, which was in agreement with the results of another report, which showed a significant decrease in the COX-2 level within the I/R kidney (Villanueva et al., 2007). Although some studies have demonstrated that COX-2 was involved in I/R pathology and its inhibition could reduce renal damage and oxidative stress (Feitoza et al., 2005; Suleyman et al., 2014; Suleyman et al., 2015), evidence has indicated that pharmacological or genetic blockage of COX-2 increased renal injury and dysfunction following renal I/R (Hwang et al., 2013; Patel et al., 2007). The results of the present study disclosed that treatment with NC did not restore the COX level in the renal tissue, as there was no significant difference between the I/R and NC + I/R groups. Therefore, it seems that the protective effect of NC is not dependent on COX-2 restoration.

The present study findings revealed an increase in renal caspase-3 cleavage following I/R. Caspase-3 is considered the main effector of apoptosis (McComb et al., 2019) and its upregulation has been reported following renal I/R injury (Karimi et al., 2021; Yang et al., 2018). In the current study, NC significantly reduced caspase-3 cleavage in the I/R animals. Thus, it could be concluded that the inhibition of apoptosis was involved in the protective effect of NC in renal I/R. This finding was compatible with those of the previous studies showing the anti-apoptotic impact of curcuminoid nanoparticles in the human renal tubular epithelial cell line (Xu et al., 2016) as well as in the renal tissue (Karimi et al., 2021).

In conclusion, the present study results indicated that NC had renoprotective and anti-apoptotic effects in the rat model of renal I/R. Moreover, renal I/R led to COX-2 and Na^+/K^+-ATPase downregulation, while the nephroprotective effect of NC was not dependent on $Na^+/$

K^+-ATPase and COX-2 restoration.

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Conflict of Interest

The authors declare that they have no conflict of interest.

Ethical statement

The protocols were approved by the Animal Experiment Ethics Committee of Shiraz University of Medical Sciences, Shiraz, Iran (license # IR.SUMS.REC.1399.556). The study did not contain any experiments on human participants performed by any of the authors.

References

Amalraj A, Pius A, Gopi S, Gopi S. Biological activities of curcuminoids, other biomolecules from turmeric and their derivatives - A review. J Tradit Complement Med 2016; 7: 205-33. https://doi.org/10.1016/j.jtcme.2016.05.005.

Amiri E, Ghasemi R, Moosavi M. Agmatine protects against 6-OHDA-induced apoptosis, and ERK and Akt/GSK disruption in SH-SY5Y cells. Cell Mol Neurobiol 2016; 36: 829-38. https://doi.org/10.1007/s10571-015-0266-7.

Aydin M S, Caliskan A, Kocarslan A, Kocarslan S, Yildiz A, Günay S, et al. Intraperitoneal curcumin decreased lung, renal and heart injury in abdominal aorta ischemia/reperfusion model in rat. Int J Surg 2014; 12: 601-5. https://doi.org/10.1016/j.ijsu.2014.04.013.

Clausen MV, Hilbers F, Poulsen H. The structure and function of the na,k-atpase isoforms in health and disease. Front physiol 2017; 8: 371. https://doi.org/10.3389/fphys.2017.00371.

Dawidczyk CM, Kim C, Park JH, Russell LM, Lee KH, Pomper MG, et al. State-of-the-art in design rules for drug delivery platforms: lessons learned from FDA-approved nanomedicines. J Control Release 2014; 187: 133-44. https://doi.org/10.1016/j.jconrel.2014.05.036.

Feitoza CQ, Câmara NO, Pinheiro HS, Gonçalves GM, Cenedeze MA, Pacheco-Silva A, et al. Cyclooxygenase 1 and/or 2 blockade ameliorates the renal tissue damage triggered by ischemia and reperfusion injury. Int Immunopharmacol 2005; 5: 79-84. https://doi.org/10.1016/j.intimp.2004.09.024.

Féraille E, Doucet A. Sodium-potassium-adenosinetriphosphatase-dependent sodium transport in the kidney: hormonal control. Physiol Rev 2001; 81: 345-418. https://doi.org/10.1152/physrev.2001.81.1.345.

Gholampour F, Roozbeh J, Janfeshan S, Karimi Z. Remote ischemic per-conditioning protects against renal ischemia-reperfusion injury via suppressing gene expression of TLR4 and TNF-alpha in rat model. Can J Physiol Pharmacol 2019; 97: 112-9. https://doi.org/10.1139/cjpp-2018-0543.

Goetz Moro M, Vargas Sánchez PK, Lupepsa AC, Baller EM, Nobre Franco GC. Cyclooxygenase biology in renal function - literature review. Revista Colombiana de Nefrología 2017; 4: 27-37. https://doi.org/10.22265/acnef.4.1.263.

Hatamipour M, Sahebkar A, Alavizadeh SH, Dorri M, Jaafari MR. Novel nanomicelle formulation to enhance bioavailability and stability of curcuminoids. Iran J Basic Med Sci 2019; 22: 282-9.

Hwang HS, Yang KJ, Park KC, Choi HS, Kim SH, Hong SY, et al. Pretreatment with paricalcitol attenuates inflammation in ischemia-reperfusion injury via the up-regulation of cyclooxygenase-2 and prostaglandin E2. Nephrol Dial Transplant 2013; 28: 1156-66. https://doi.org/10.1093/ndt/gfs540.

Kar F, Hacioglu C, Senturk H, Donmez DB, Kanbak G, Uslu S. Curcumin and LOXblock-1 ameliorate ischemia-reperfusion induced inflammation and acute kidney injury by suppressing the semaphorin-plexin pathway. Life Sci 2020; 256: 118016. https://doi.org/10.1016/j.lfs.2020.118016.

Karahan MA, Yalcin S, Aydogan H, Büyükfirat E, Kücük A, Kocarslan S, et al. Curcumin and dexmedetomidine prevents oxidative stress and renal injury in hind limb ischemia/reperfusion injury in a rat model. Renal Failure 2016; 38: 693-8. https://doi.org/10.3109/0886022X.2016.1157746.

Karimi Z, SoukhakLari R, Rahimi-Jaberi K, Esmaili Z, Moosavi M. Nanomicellar curcuminoids attenuates renal ischemia/reperfusion injury in rat through prevention of apoptosis and downregulation of MAPKs pathways. Mol Biol Rep 2021; 48: 1735-43. https://doi.org/10.1007/s11033-021-06214-2.

Kaur A, Kaur T, Singh B, Pathak D, Singh Buttar H, Pal Singh A. Curcumin alleviates ischemia reperfusion-induced acute kidney injury through NMDA receptor antagonism in rats.

Ren Fail 2016; 38: 1462-7. https://doi.org/10.1080/0886022X.2016.1214892.

Liu F, Ni W, Zhang J, Wang G, Li F, Ren W. Administration of curcumin protects kidney tubules against renal ischemia-reperfusion injury (RIRI) by modulating nitric oxide (NO) signaling pathway. Cell Physiol Biochem 2017; 44: 401-11. https://doi.org/10.1159/000484920.

Liu FH, Ni WJ, Wang GK, Zhang JJ. Protective role of curcumin on renal ischemia reperfusion injury via attenuating the inflammatory mediators and Caspase-3. Cell Mol Biol 2016a; 62: 95-9.

Liu W, Zhai Y, Heng X, Che FY, Chen W, Sun D, et al. Oral bioavailability of curcumin: problems and advancements. J Drug Target 2016b; 24: 694-702. https://doi.org/10.3109/1061186X.2016.1157883.

Lugo-Baruqui JA, Ayyathurai R, Sriram A, Pragatheeshwar KD. Use of mannitol for ischemia reperfusion injury in kidney transplant and partial nephrectomies-review of literature. Curr Urol Rep 2019; 20: 6. https://doi.org/10.1007/s11934-019-0868-6.

Malek M, Nematbakhsh M. Renal ischemia/reperfusion injury; from pathophysiology to treatment. J Ren Inj Prev 2015; 4: 20-7.

Matsuzaki T, Watanabe H, Yoshitome K, Morisaki T, Hamada A, Nonoguchi H, et al. Downregulation of organic anion transporters in rat kidney under ischemia/reperfusion-induced acute [corrected] renal failure. Kidney Int 2007; 71: 539-47. https://doi.org/10.1038/sj.ki.5002104.

McComb S, Chan PK, Guinot A, Hartmannsdottir H, Jenni S, Dobay MP, et al. Efficient apoptosis requires feedback amplification of upstream apoptotic signals by effector caspase-3 or -7. Sci Adv 2019; 5: 9433. https://doi.org/10.1126/sciadv.aau9433

Moosavi M, Abbasi L, Zarifkar A, Rastegar K. The role of nitric oxide in spatial memory stages, hippocampal ERK and CaMKII phosphorylation. Pharmacol Biochem Behav 2014; 122: 164-72. https://doi.org/10.1016/j.pbb.2014.03.021

Najafi H, Changizi Ashtiyani S, Sayedzadeh SA, Mohamadi Yarijani Z, Fakhri S. Therapeutic effects of curcumin on the functional disturbances and oxidative stress induced by renal ischemia/reperfusion in rats. Avicenna J Phytomed 2015; 5: 576-86.

Nørregaard R, Kwon T-H, Frøkiær J. Physiology and pathophysiology of cyclooxygenase-2 and prostaglandin E2 in the kidney. Kidney Res Clin Pract 2015; 34: 194-200. https://doi.org/10.1016/j.krcp.2015.10.004.

Patel NS, Cuzzocrea S, Collino M, Chaterjee PK, Mazzon E, Britti D, et al. The role of cycloxygenase-2 in the rodent kidney following ischaemia/reperfusion injury in vivo. Eur J Pharmacol 2007; 562: 148-54. https://doi.org/10.1016/j.ejphar.2007.01.079

Prasad S, Tyagi AK, Aggarwal BB. Recent developments in delivery, bioavailability, absorption and metabolism of curcumin: the golden pigment from golden spice. Cancer Res Treat 2014; 46: 2-18. https://doi.org/10.4143/crt.2014.46.1.2.

Sampaio LS, Iannotti FA, Veneziani L, Borelli-Tôrres RT, De Maio F, Piscitelli F, et al. Experimental ischemia/reperfusion model impairs endocannabinoid signaling and Na^+/K^+ ATPase expression and activity in kidney proximal tubule cells. Biochem Pharmacol 2018; 154: 482-91. https://doi.org/10.1016/j.bcp.2018.06.005.

Shiva N, Sharma N, Kulkarni YA, Mulay SR, Gaikwad AB. Renal ischemia/reperfusion injury: An insight on in vitro and in vivo models. Life Sci 2020: 117860. https://doi.org/10.1016/j.lfs.2020.117860.

Soares ROS, Losada DM, Jordani MC, Évora P, Castro-E-Silva O. Ischemia/Reperfusion injury revisited: an overview of the latest pharmacological strategies. Int J Mol Sci 2019; 20: 5034. https://doi.org/10.3390/ijms20205034.

Suleyman B, Albayrak A, Kurt N, Demirci E, Gundogdu C, Aksoy M. The effect of etoricoxib on kidney ischemia-reperfusion injury in rats: A biochemical and immunohistochemical assessment. Int Immunopharmacol 2014; 23: 179-85. https://doi.org/10.1016/j.intimp.2014.06.042.

Suleyman Z, Sener E, Kurt N, Comez M, Yapanoglu T. The effect of nimesulide on oxidative damage inflicted by ischemia-reperfusion on the rat renal tissue. Ren Fail 2015; 37: 323-31. https://doi.org/10.3109/0886022X.2014.985996.

Villanueva S, Céspedes C, González AA, Vio CP, Velarde V. Effect of ischemic acute renal damage on the expression of COX-2 and oxidative stress-related elements in rat kidney. Am J Physiol Renal Physiol 2007; 292: 1364-71. https://doi.org/10.1152/ajprenal.00344.2006.

Williams HD, Trevaskis NL, Charman SA, Shanker RM, Charman WN, Pouton CW, et al. Strategies to address low drug solubility in discovery and development. Pharmacol Rev 2013; 65: 315-499. https://doi.org/10.1124/pr.112.005660.

Wright J, Healy T, Balfour T, Hardcastle J. Effects of inhalation anaesthetic agents on the electrical and mechanical activity of the rat duodenum. Br J Anaesth 1982; 54: 1223-30. https://doi.org/10.1093/bja/54.11.1223.

Xu Y, Hu N, Jiang W, Yuan HF, Zheng DH. Curcumin-car-

rying nanoparticles prevent ischemia-reperfusion injury in human renal cells. Oncotarget 2016; 7: 87390-401. https://doi.org/10.18632/oncotarget.13626.

Yang B, Lan S, Dieudé M, Sabo-Vatasescu J-P, Karakeussian-Rimbaud A, Turgeon J, et al. Caspase-3 Is a pivotal regulator of microvascular rarefaction and renal fibrosis after ischemia-reperfusion injury. J Am Soc Nephrol 2018; 29: 1900-16. https://doi.org/10.1681/ASN.2017050581.

Zhang J, Tang L, Li GS. The anti-inflammatory effects of curcumin on renal ischemia-reperfusion injury in rats. 2018; 40: 680-6. https://doi.org/10.1080/0886022X.2018.1544565.