Paricalcitol may improve oxidative DNA damage on experimental amikacin-induced nephrotoxicity model

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**ABSTRACT**

This study aimed to investigate the possible protective effect of paricalcitol on experimental amikacin-induced nephrotoxicity model in rats. Wistar albino rats ($n = 32$) were allocated into four equal groups of eight each, the control (Group C), paricalcitol (Group P), amikacin-induced nephrotoxicity (Group A), and paricalcitol-treated amikacin-induced nephrotoxicity (Group A + P) groups. Paricalcitol was given intra-peritoneally at a dose of 0.4 μg/kg/d for 5 consecutive days prior to induction of amikacin-induced nephrotoxicity. Intra-peritoneal amikacin (1.2 g/kg) was used to induce nephrotoxicity at day 4. Renal function parameters, oxidative stress biomarkers, oxidative DNA damage (8-hydroxy-2'-deoxyguanosine/deoxyguanosine ratio), kidney histology, and vascular endothelial growth factor (VEGF) immunoexpression were determined. Group A + P had lower mean fractional sodium excretion ($p < 0.001$) as well as higher creatinine clearance ($p = 0.026$) than the amikacin group (Group A). Renal tissue malondialdehyde levels ($p = 0.035$) and serum 8-hydroxy-2'-deoxyguanosine/deoxyguanosine ratio (8-OHdG/dG ratio) ($p < 0.001$) were significantly lower; superoxide dismutase ($p = 0.024$) and glutathione peroxidase ($p = 0.007$) activities of renal tissue were significantly higher in group A + P than in group A. The mean scores of tubular necrosis ($p = 0.024$), proteinaceous casts ($p = 0.038$), medullary congestion ($p = 0.035$), and VEGF immunoexpression ($p = 0.018$) were also lower in group A + P when compared with group A. This study demonstrates the protective effect of paricalcitol in the prevention of amikacin-induced nephrotoxicity in an experimental model. Furthermore, it is the first study to demonstrate that paricalcitol improves oxidative DNA damage in an experimental acute kidney injury model.

**Introduction**

Aminoglycosides are among the oldest antibiotics available to treat serious infections caused by primarily Gram-negative bacteria. As the use of aminoglycosides became more widespread, the toxic effects of these agents, most notably ototoxicity and nephrotoxicity, became more apparent. Intracellular accumulation of the drug in the proximal tubules may lead to oxidative stress, tubular toxicity, and cell necrosis.\textsuperscript{1} Recovery from drug-induced nephrotoxicity is usually slow, particularly in elderly individuals.\textsuperscript{2} Patients with underlying chronic kidney disease may experience incomplete recovery of renal function.\textsuperscript{2}

Paricalcitol (19-nor-1,25-dihydroxyvitamin D\textsubscript{2}) is an active, non-hypercalcemic vitamin D analog that shows similar biological activity with vitamin D, but has fewer adverse effects.\textsuperscript{3} In addition to its primary role in calcium metabolism and bone mineralization, vitamin D and its non-hypercalcemic analog paricalcitol have pleiotropic and anti-oxidant effects on cellular homeostasis.\textsuperscript{4} Studies in experimental nephropathy models have focused on the effects of vitamin D and paricalcitol on glomerular damage and tubular toxicity.\textsuperscript{5–7} Recently, it has been shown that paricalcitol has anti-oxidant effects on renal tissue and suppresses the renin-angiotensin system (RAS) in diabetic kidney.\textsuperscript{8,9} Fryer et al.\textsuperscript{10} demonstrated that paricalcitol suppresses renal renin expression. It has also been recently reported that the combination of paricalcitol and enalapril treatment ameliorates oxidative injury by suppressing reactive oxygen species generating enzyme NADPH oxidase activity and by up regulating the antioxidant defense system in a uremic rat model of atherosclerosis.\textsuperscript{11} It is not known, however, whether paricalcitol can protect against oxidative stress in amikacin-induced nephrotoxicity.

The aim of the present study was to investigate the effect of paricalcitol in the experimental amikacin-induced nephrotoxicity model in rats.
We hypothesized that paricalcitol may prevent amikacin-induced nephrotoxicity due to its anti-oxidant effects in the kidney. We evaluated renal function parameters, intra-renal levels, and activities of oxidative stress biomarkers and renal histology after treatment with paricalcitol.

Methods

Animals

The experimental procedures were performed in accordance with the guide for the Care and Use of Laboratory Animals (National Institutes of Health) and were approved by the Institutional Animal Care Committee of Yuzuncu Yil University. The study included 32 male Wistar Albino rats (12 weeks old), each weighing 250–280 g, which were born and bred in the Experimental Research Center of Yuzuncu Yil University. They were kept in stainless steel cages, maintained on a 12-h-light/12-h-dark cycle at 22–25°C where they had unlimited access to standard rat chow and water. Individual metabolic cages were used for the collection of 24 h urine and for recording water intake.

Experimental design and drugs

The animals were randomly allocated into four groups:

Control group (Group C) (n = 8): Control animals received intraperitoneal (i.p.) saline injections daily, for 5 consecutive days.

Paricalcitol group (Group P) (n = 8): Rats received paricalcitol (Zemplar, Abbott, IL) i.p. at a dose of 0.4 μg/kg/d once daily for 5 consecutive days.8

Amikacin group (Group A) (n = 8): Amikacin (Amikozit 500 mg, Eczacibasi Corp., Istanbul, Turkey) at a dose of 1.2 g/kg was injected intraperitoneally at day 4.12,13

Paricalcitol + amikacin group (Group A + P) (n = 8): In this group, rats received paricalcitol i.p. at a dose of 0.4 μg/kg/d once daily for 5 consecutive days. Amikacin was applied to this group as described for group A.

Rats were allowed to recover in the metabolic cages for an additional 24 h-period at the end of which, 24 h urine samples were collected for creatinine clearance (CCr) and fractional Na clearance (FENa%) measurement. Blood samples were withdrawn from the abdominal aorta and the right kidneys were excised under general anesthesia, achieved by i.p. injections of ketamine (Ketalar, Pfizer, Turkey) at 50 mg/kg. Kidney, blood, and urine samples were taken, serum was separated and aliquots were stored at −80°C until analysis.

Evaluation of renal functions

Serum and urinary creatinine and sodium were measured using commercially available clinical assay kits with an autoanalyzer (COBAS Integra 400 Plus, Roche Diagnostic, Rotkreuz, Switzerland) according to the instructions of the manufacturers. CCr was calculated using the following equation:

$$\text{CCr (milliliters per minute per kilogram of body weight)} = \left[ \frac{\text{urinary Cr (milligrams per deciliter)}}{\text{urinary volume (mL)/serum Cr (milligrams per deciliter)}} \right] \times \left[ 1000/\text{body weight (g)} \right] \times 1/1440 \text{ (min)}.14$$

Fractional excretion of sodium (FENa) was calculated as the following: (urine sodium/serum sodium)/(serum creatinine/urine creatinine) × 100.

Oxidative stress biomarkers

Determination of MDA

Measurement of plasma malondialdehyde (MDA) concentration was performed according to Khoschsorur et al.15 Briefly, 50 μL of plasma sample was mixed with 0.44 M H3PO4 and 42 mM thiobarbituric acid (TBA) and incubated for 30 min in a boiling water bath. After rapidly cooling on ice, an equal volume of alkaline methanol was added to the sample, vigorously shaken, centrifuged (3000 rpm for 3 min), and the aqueous layer was removed. Then, 20 μL of supernatant was analyzed by HPLC (HP, Agilent 1100 modular systems with FLD detector, Agilent, Waldbronn, Germany): column, RP-C18 (5 μm, 4.6 × 150 mm, Eclipse VDB-C18, Agilent, Waldbronn, Germany); elution, methanol (40:60, v/v) containing 50 mM KH2PO4 buffer (pH 6.8); flow rate, 0.8 mL/min. Fluorometric detection was performed with excitation at 527 nm and emission at 551 nm. The peak of the MDA-TBA adduct was calibrated as a 1,1,3,3-tetraethoxypropane standard solution carried out in exactly the same process as with the plasma sample.

Determination of DNA Damage

Fasting blood samples were obtained from all subjects and collected into tubes without coagulant. Serum was obtained by centrifugation at 2500 rpm for 15 min and stored at −80°C until assayed.

Isolation and hydrolyzation of DNA

DNA isolation from whole blood was performed according to Miller et al.16 with some modifications.
Two milliliter of blood with ethylene diamine tetraacetic acid (EDTA) was mixed with 3 mL of erythrocyte lysis buffer, and incubation for 10 min in ice was followed by centrifugation (10 min at 3500 rpm). The supernatant was decanted, and the pellet was thoroughly resuspended in sodium dodecyl sulfate (10%, v/v), proteinase K (20 mg/mL), and 1.9 mL leukocyte lysis buffer. The mixture was incubated at 65 °C for 1 h and then mixed with 0.8 mL of 9.5 M ammonium acetate. After centrifugation at 3500 rpm for 25 min, the clear supernatant (2 mL) was transferred to a new sterile tube, and DNA was precipitated by addition of 4 mL of ice-cold absolute ethanol. DNA samples were dissolved in Tris EDTA buffer (10 mM, pH 7.4), and then were hydrolyzed according to Shigenaga’s method.

**Analysis of 8-OHdG and dG by the HPLC method**

In the hydrolyzed DNA samples, 8-OHdG and dG levels were measured, respectively, by HPLC with electrochemical (HPLC-ECD) and variable wavelength detector (HPLC-UV) systems as previously described. Twenty microliter of final hydrolysate were analyzed by HPLC-ECD (HP, Agilent 1100 modular systems with HP 1049A ECD detector, Agilent, Waldbronn, Germany): column, reverse phase-C18 (RP-C18) analytical column (250 mm x 4.6 mm x 4.0 μm, Phenomenex, Torrance, CA). The mobile phase consisted of 0.05 M potassium phosphate buffer [pH 5.5] containing acetonitrile (97: 3, v/v) with a flow rate of 1 mL/min. The dG concentration was monitored based on the absorbance (245 nm) and 8-OHdG based on the electrochemical reading (600 mV). Levels of dG and 8-OHdG were quantified using the standards of dG and 8-OHdG from Sigma (St. Louis, MO); the level of 8-OHdG is expressed as the number of 8-OHdG molecules per 106 dG.

**Determination of the antioxidant activity**

The homogenization of tissues was carried out in a Teflon-glass homogenizer with a buffer containing 1.15% KCl to obtain 1/10 (w/v) whole homogenate. The homogenates were centrifuged 30 min at 25,000 g at +4 °C to determine glutathione peroxidase (GSH-Px) activity. The GSH-Px activities were determined according to the method of Beutler which records the disappearance of NADPH at 340 nm, and results were expressed as U/g protein. The obtained supernatants were centrifuged again at 25,000 g, +4 °C for 30 min to determine superoxide dismutase (SOD) activities. SOD activities were determined using the method of Sun et al. and the results were expressed as U/g protein.

**Histology**

The excised kidneys were preserved in phosphate-buffered 10% formalin, embedded in paraffin wax, and cut into 3 μm sections according to conventional techniques. The sections were stained with hematoxylin–eosin. Histopathological changes were analyzed for tubular necrosis, proteinaceous casts, and medullary congestion, as suggested by Solez et al. Tubular necrosis and proteinaceous casts were graded as follows: no damage (0), mild (+1, unicellular, patchy isolated damage), moderate (+2, damage less than 25%), severe (+3, damage between 25 and 50%), and very severe (+4, more than 50% damage). The degree of medullary congestion was defined as no congestion (0), mild (+1 vascular congestion with identification of erythrocytes by 400× magnification), moderate (+2 vascular congestion with identification of erythrocytes by 200× magnification), severe (+3 vascular congestion with identification of erythrocytes by 100× magnification), and very severe (+4 vascular congestion with identification of erythrocytes by 40× magnification). Evaluations were made by an experienced pathologist who was blind to the data.

**Immunohistochemistry for VEGF**

Immunohistochemistry for VEGF was performed on 3-μm-thick renal sections based on streptavidin–biotin–peroxidase complex formation, according to the instructions of the manufacturer. In brief, paraffin-embedded sections were cleared in xylene, rehydrated in a series of ethanol washes. Endogenous peroxidase activity was inhibited with 3% H2O2. Antigen retrieval was performed by microwaving sections in citrate buffer (pH: 6). Sections were blocked in PBS (pH: 7.4) for 20 min at room temperature and then protein blockage was performed for inhibition of non-specific staining. Sections were incubated with anti-VEGF (Ab-1, RB-222-R7; Thermo Fisher Scientific, Fremont, CA) for 30 min, then incubated with biotinylated secondary antibody (UltraVision Detection System; TP-015-HD; Thermo Fisher Scientific, Fremont, CA) for 10 min. Finally streptavidin peroxidase (UltraVision Detection System; TP-015-HD; Thermo Fisher Scientific, Fremont, CA) was added for 10 min, and sections were washed in PBS (ph:7.4) before detection with DAB reagent. After briefly being counterstained with Mayer’s hematoxylin, sections were dehydrated. Negative control sections were stained under identical conditions by
omitting the primary antibody. Human colon cancer tissue was used as a positive control in staining.

To assess immunohistological staining, tubular VEGF immunostaining was assessed semiquantitatively in the outer medulla of the kidney (nuclear staining of the tubular cells): very weak or absent nuclear stain (0), weak nuclear stain of <25% of tubular cells (1), 25–50% of tubular cells were stained (2), 50–75% of tubular cells were stained (3), and >75% of tubular cells were stained intensely (4).

### Statistical analysis

Data are presented as mean ± SD. ANOVA and Bonferroni’s tests were used to compare the means of continuous variables that were normally distributed and had equal variances. Welch and Games–Howel tests were used to compare the means of continuous variables that were normally distributed and did not have equal variances. The Kruskal–Wallis and Mann–Whitney U tests were used to compare the medians of non-normally distributed continuous variables. Two-tailed p values <0.05 were considered statistically significant. Statistical analyses were performed using SPSS version 19.0 (SPSS Inc., Chicago, IL).

### Results

#### Renal function parameters

Table 1 shows the renal function parameters in the four treatment groups. Twenty-four hour CCr did not differ in the C, P, and A + P groups. On one hand, as expected, CCr decreased significantly in group A when compared with control animals (p < 0.005). CCr was significantly higher in group A + P than in group A (p = 0.026), but no differences were seen in group A + P when compared with control rats (p > 0.05). The mean FENa did not differ in the C, P, and A + P groups. FENa was significantly higher in group A than in controls (p < 0.005). On the other hand, this parameter was significantly lower in group A + P than in group A (p < 0.001) (Table 1).

#### Oxidative stress parameters

Table 2 shows the changes in oxidative stress biomarkers in the four treatment groups. Statistical analysis showed that the mean kidney MDA levels were significantly lower in the group A + P than in group A (p = 0.035), but there were no differences between group A + P and either group P or control group (p > 0.05). Similarly, oxidative DNA damage (8-OHdG/dG ratio) was increased in Group A when compared with the control group (p < 0.005). Oxidative DNA damage (8-OHdG/dG ratio) was significantly lower in group A + P than in group A (p < 0.001), but no differences were seen in group A + P when compared with control rats (p > 0.05). SOD (p = 0.024) and GSH-Px (p = 0.007) activities were significantly higher in group A + P than in group A. There were no significant differences of these parameters in Group A + P when compared with controls (p > 0.05) (Table 2).

#### Histological and immunohistochemical evaluation

##### Histological analysis

The histological findings in the four treatment groups are reported in Table 3. The scores of tubular necrosis (p < 0.005), proteinaceous casts (p < 0.005), and medullary congestion (p < 0.005) parameters were significantly higher in group A + P than in group A + P when compared with controls (p > 0.05) (Table 2).

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**Table 1. Renal function parameters in the four treatment groups (n = 8 each).**

|                      | Control group (group C) | Amikacin group (group A) | Paricalcitol + amikacin group (group A + P) | Paricalcitol group (group P) |
|----------------------|-------------------------|--------------------------|---------------------------------------------|-----------------------------|
| Creatinine clearance (mL/min/mg) | 8.32 ± 1.24            | 4.96 ± 0.75              | 6.48 ± 0.69<sup>a</sup>                     | 8.04 ± 1.06                 |
| Fractional excretion of sodium (%) | 0.20 ± 0.10            | 1.74 ± 0.44              | 0.97 ± 0.40<sup>b</sup>                     | 0.41 ± 0.02                 |

Data are presented as mean ± standard deviation.

<sup>a</sup>p = 0.026, group A + P versus group A (ANOVA followed by Mann–Whitney U test). <sup>b</sup>p < 0.001, group A + P versus group A (ANOVA followed by Mann–Whitney U test).

**Table 2. Oxidative stress biomarkers of four treatment groups (n = 8 each).**

|                      | Control group (group C) | Amikacin group (group A) | Paricalcitol + amikacin group (group A + P) | Paricalcitol group (group P) |
|----------------------|-------------------------|--------------------------|---------------------------------------------|-----------------------------|
| MDA (nmol/g protein) | 16.74 ± 3.18            | 34.47 ± 6.08             | 21.18 ± 4.96<sup>a</sup>                    | 17.04 ± 2.60                |
| 8-OHdG/DG            | 0.63 ± 0.48             | 2.49 ± 1.14              | 0.94 ± 0.34<sup>b</sup>                     | 0.65 ± 0.07                 |
| SOD (U/g protein)    | 48.80 ± 9.22            | 31.41 ± 12.14            | 40.05 ± 7.24<sup>a</sup>                    | 47.19 ± 10.62               |
| GSH-Px (U/g protein) | 32.16 ± 7.74            | 19.08 ± 6.04             | 28.45 ± 5.84<sup>b</sup>                    | 32.02 ± 5.92                |

MDA: malondialdehyde; 8-OHdG/DG: 8-hydroxy deoxyguanosine/deoxyguanosine; SOD: superoxide dismutase; GSH-Px: glutathione peroxidase. Data are presented as mean ± standard deviation.

<sup>a</sup>p < 0.05, group A + P versus group A (ANOVA followed by Mann–Whitney U test). <sup>b</sup>p < 0.001, group A + P versus group A (ANOVA followed by Mann–Whitney U test).
different between control and amikacin-induced nephrotoxicity groups. Tubular necrosis ($p = 0.024$), proteinaceous casts ($p = 0.038$), and medullary congestion ($p = 0.035$) parameters were significantly lower in group A + P than in group A. However, there were no significant differences of these parameters in Group A + P when compared with controls ($p > 0.05$, respectively) (Figure 1).

**Immunohistochemistry**

Immunohistochemical analysis of VEGF in the tubular cells was significantly increased in group A when compared with controls ($p < 0.005$). The mean VEGF score was significantly lower in group A + P than in group A ($p = 0.018$), but no difference was seen in group A + P when compared to control rats ($p > 0.05$) (Figure 2).

**Table 3.** Histology and immunohistochemistry in the four treatment groups ($n = 8$ each).

|                      | Amikacin group (group A) | Paricalcitol + amikacin group (group A + P) | Paricalcitol group (group P) |
|----------------------|---------------------------|---------------------------------------------|-----------------------------|
| Tubular necrosis     | 0.64 ± 0.76               | 1.47 ± 0.51a                               | 0.72 ± 0.89                 |
| Proteinaceous cast   | 0.55 ± 0.48               | 1.96 ± 1.28a                               | 0.46 ± 0.74                 |
| Medullary congestion | 0.48 ± 0.35               | 1.87 ± 0.64a                               | 0.52 ± 0.70                 |
| VEGF score           | 0.50 ± 0.52               | 1.25 ± 0.53a                               | 0.54 ± 0.25                 |

VEGF: vascular endothelial growth factor. Data are presented as mean ± standard deviation. $^a p < 0.05$, group A + P versus group A (Kruskal–Wallis followed by Mann–Whitney $U$ test).

**Discussion**

In the present experimental study, we have demonstrated for the first time that prophylaxis using paricalcitol protects against acute kidney injury in the amikacin-induced nephrotoxicity model in rats. Paricalcitol decreased intra-renal oxidative stress and systemic oxidative DNA damage, reduced tubular necrosis, medullary congestion, and proteinaceous casts which had occurred secondary to amikacin administration and lowered VEGF immunoexpression that had presumably developed secondary to regional renal hypoxia.

Aminoglycoside-induced nephrotoxicity is a relatively common complication in hospitalized patients that leads to significant morbidity and additional cost of therapy. Aminoglycosides are not metabolized in the

![Figure 1](image1.png)  
**Figure 1.** Renal histology of the study groups (haematoxylin and eosin staining, original magnification, x400). Normal histology in (a) control and (c) paricalcitol groups. (b) Tubular necrosis areas and proteinaceous casts of Group A. (d) Healthy tubular cells of Group A + P.
body and most of the injected dose is excreted in the urine, whereas drug concentration in renal proximal tubular cells is several times higher than that of the plasma concentration. Furthermore, the concentration of the drug in the proximal tubular cells is associated with its nephrotoxic effects. Accumulated amikacin in the renal proximal tubular cells generate free radicals that induce an increase in oxidative stress. In the present study, the activity of SOD and GSH-Px in glomeruli of injury rats was evidently inhibited along with a distinct increase in MDA level. In contrast, following paricalcitol treatment, the activity of all the enzymes displayed a significant increase accompanied with the obvious decrease in MDA level. Ulusoy et al. reported that MDA, total oxidative system, and oxidative stress indexes increase on experimental amikacin-induced nephrotoxicity model. Similarly Kose et al. demonstrated that intra-renal levels of MDA and myeloperoxidase increase in amikacin-induced toxicity in rats. Our results demonstrate that paricalcitol treatment possesses antioxidant activity and could improve the antioxidative defense mechanism in acute kidney injury. We demonstrated that paricalcitol blocks cell injury during aminoglycoside-induced toxicity through reduction in cellular oxidative stress.

As oxidative stress has been implicated in the pathogenesis of amikacin-induced nephrotoxicity, there is growing evidence for the presence of disordered oxidative chemistry in amikacin-induced acute kidney injury experimental models which may contribute to poor renal outcome. DNA, in particular, is more susceptible to attack by reactive oxygen species than proteins and membrane lipids, which are protected by low-molecular weight anti-oxidants and anti-oxidant enzymes. Among many types of oxidative DNA damage, 8-OHdG is one of the most abundant oxidative products of cellular DNA. According to our results, paricalcitol prevents not only intra-renal oxidative stress but also systemic oxidative DNA damage. There is no data regarding cellular DNA damage in aminoglycoside nephrotoxicity or the protective effect of paricalcitol on cellular DNA in this setting of acute kidney injury in the literature. Akt-signaling pathway is known to be involved in the survival-signaling pathway. Previous reports demonstrated that the activation of Akt-signaling pathways is associated with attenuation of apoptosis in the kidney against various noxious stimuli. Although Akt-signaling pathway or apoptosis mechanisms were not evaluated in our study, it is postulated that Akt-signaling pathway was activated by amikacin as

Figure 2. Immunoexpression of VEGF in the four treatment groups (X400). No VEGF expression in the renal tubular cells in the (a) control, (c) paricalcitol and (d) paricalcitol treated amikacin-induced nephrotoxicity groups. (b) Intense expression of VEGF of amikacin-induced nephrotoxicity group.
a defense mechanism. Paricalcitol blocks renal tubular injury and renal apoptosis by reducing oxidative stress, oxidative DNA damage, and probably the activity of Akt signaling in mice. Suh et al. reported that gentamicin increases caspase-3 activity and Jun-N-terminal kinase expression in rat kidney, which was counteracted by paricalcitol. Although we could not study these apoptotic mechanisms, it is reasonable to interpret that paricalcitol may have anti-apoptotic effects. Beyond the hypothetical mechanisms, this study provides, for the first time, evidence that oxidative DNA damage, in terms of 8-OHdG/dG ratio, does exist in aminoglycoside toxicity and paricalcitol improves oxidative DNA damage in this setting of acute kidney injury. We believe that this finding may have important implications for new therapeutic interventions in amikacin-induced nephrotoxicity.

Izquierdo et al. reported that paricalcitol reduces oxidative stress and inflammation in hemodialysis patients. Furthermore, paricalcitol has been shown to attenuate contrast-induced nephropathy by suppression of renal and systemic oxidative stress. Eren et al. also recently reported that paricalcitol improves albuminuria and intrarenal oxidative stress in diabetic rats. These findings that address the pleiotropic effects of paricalcitol and our findings suggest that paricalcitol may well have benefits in the chronic as well as acute settings.

The observed benefits of paricalcitol administration can further be associated with the inhibition of tubular expression of VEGF, even though the precise mechanisms by which paricalcitol ameliorates VEGF expression remains to be studied. Freundlich et al. reported that paricalcitol treatment reduces mRNA and protein expression of VEGF in the kidney in the experimental chronic renal failure model, possibly through the inhibition of RAS. Ping et al. demonstrated that xenon pretreatment upregulates hypoxia-inducible factor-2α and its downstream effector VEGF in experimental aminoglycoside nephrotoxicity. In our study, acute inhibition of VEGF overexpression by paricalcitol might be related to the improvement of renal and systemic oxidative stress and subsequent amelioration of local renal hypoxia. Future studies are needed to shed more light on the potential pathophysiological effects of paricalcitol on tubular VEGF immunoexpression on aminoglycoside nephrotoxicity.

In conclusion, the present study has revealed a novel effect of the active vitamin D analog paricalcitol in protection against amikacin-induced nephrotoxicity, possibly through its anti-oxidant effects by inhibition of lipid peroxidation and oxidative DNA damage. Further experimental studies and clinical trials on paricalcitol would be valuable in providing further insight into paricalcitol renoprotection against aminoglycoside nephrotoxicity and its clinical application.

Disclosure statement
The authors report that they have no conflicts of interest. The authors alone are responsible for the content and writing of the paper.

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