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SUMOylation occurs in acute kidney injury and plays a cytoprotective role

Chunyuan Guo [a], Qingqing Wei [a], Yunchao Su [c], Zheng Dong [a,b,d,*]

[a] Department of Cellular Biology and Anatomy, Georgia Regents University, Augusta, GA 30912, United States
[b] Charlie Norwood VA Medical Center, Augusta, GA 30912 United States
[c] Department of Pharmacology & Toxicology, Medical College of Georgia, Georgia Regents University, Augusta, GA 30912, United States
[d] Department of Nephrology, The Second Xiangya Hospital, Central South University, Changsha, Hunan, China

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ABSTRACT

SUMOylation is a form of post-translational modification where small ubiquitin-like modifiers (SUMO) are covalently attached to target proteins to regulate their properties. SUMOylation has been demonstrated during cell stress and implicated in cellular stress response. However, it is largely unclear if SUMOylation contributes to the pathogenesis of kidney diseases, such as acute kidney injury (AKI). Here we have demonstrated a dynamic change of protein SUMOylation in ischemic and cisplatin nephrotoxic AKI in mice. In rat kidney proximal tubular cells (RPTC), cisplatin-induced SUMOylation was diminished by two antioxidants (N-acetylcysteine and dimethylurea), supporting a role of oxidative stress in the activation of SUMOylation. In addition, SUMOylation by SUMO-2/3, but not SUMO-1, was partially suppressed by pifithrin-alpha (a pharmacological inhibitor of p53), supporting a role of p53 in SUMOylation by SUMO-2/3. We further examined the role of SUMOylation during cisplatin treatment of RPTC by using ginkgolic acid (GA), a pharmacological inhibitor of SUMOylation. Pretreatment with GA suppressed SUMOylation and importantly, GA enhanced apoptosis during cisplatin incubation. Taken together, the results demonstrate the first evidence of SUMOylation in AKI and suggest that SUMOylation may play a cytoprotective role in kidney tubular cells.

1. Introduction

Acute kidney injury (AKI), formerly termed acute renal failure (ARF), is a major kidney disease associated with high mortality (>50%) [1,2]. The major causes of AKI include renal ischemia, sepsis, and nephrotoxicity [3–5]. Renal ischemia occurs during hypotension, dehydration, vascular constriction and obstruction, septic shock and operative arterial occlusion. Nephrotoxicity can be induced by environmental toxins as well as prescribed drugs such as cisplatin, one of the most commonly used drugs for cancer therapy [6–8]. Pathologically, AKI is characterized by injury and death of the cells in kidney tubules especially those in the proximal tubules [9,10]. Although the pathogenesis of AKI has been explored at various cellular and molecular levels [3,4], no information on protein SUMOylation has been reported in AKI.

SUMOylation is a reversible, post-translational modification where small ubiquitin-like modifiers (SUMO) are covalently attached to lysine residues in the target proteins [11,12]. SUMO includes a family of peptide of ~11 kDa, which consists of four isoforms: SUMO-1, -2, -3, and -4. The first three are ubiquitously expressed in all eukaryotes, while SUMO-4 is only expressed in human. The mature forms of SUMO-2 and SUMO-3 are 95% identical in sequence, thus they are often grouped together as SUMO-2/3 [11,12].

The conjugation pathway of SUMOylation is similar to that of ubiquitination, but SUMO conjugation involves a different cascade of SUMO-specific enzymes: E1—activating enzyme, E2—conjugating enzyme (Ubc9), usually one of the several SUMO E3 ligases and SUMO-specific proteolytic enzyme (SENPs) for deSUMOylation and maturation of SUMO proteins. Unlike ubiquitination, SUMOylation does not target the protein for degradation; instead it mainly regulates protein stability, localization, interaction and activity [11–13]. SUMOylation has been implicated in physiological and pathological regulations. Especially, SUMOylation has been shown to be induced by heat, hypoxic, osmotic, oxidative, genotoxic and metabolic stresses, suggesting a potential role of SUMOylation in cellular stress response [14–19].

The goal of this study is to determine the changes of protein SUMOylation in experimental models of AKI and gain some initial insights into the regulation and role of SUMOylation in this disease condition.

2. Materials and methods

2.1. Cell culture

The immortalized rat kidney proximal tubular epithelial cell (RPTC) line was obtained from Dr. Ulrich Hopfer (Case Western Reserve

* Corresponding author at: Department of Nephrology, The Second Xiangya Hospital, Central South University, Changsha, Hunan, China. Tel.: +1 706 721 2825; fax: +1 706 721 6120.
E-mail address: zdong@gru.edu (Z. Dong).

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University, Cleveland, OH) and maintained in Ham's F-12/DMEM supplemented with 10% fetal bovine serum (FBS, 5 g/ml transferrin, 5 μg/ml insulin, 1 ng/ml EGF, 4 μg/ml dexamethasone, and 1% antibiotics as previously) [20–22].

2.2. Reagents

Cisplatin, dimethyl sulfoxide (DMSO), dimethylthiourea (DMTU), nacetyl-cysteine (NAC), sodium azide, pifithrin-α and N-ethylmaleimide (NEM) were purchased from Sigma-Aldrich (St. Louis, MO). Ginkgolic acid (15:1) was purchased from EMD Millipore Corporation (Billerica, MA). Carbobenzoxy-Asp-Glu-Val-Asp-7-amino-4-trifluoromethyl coumarin (DEVD.AFC) and 7-amino-4-trifluoromethyl coumarin (AFC) were purchased from Enzyme Systems Products (Dublin, CA). Enhanced chemiluminescence kit was purchased from Pierce Biotechnology (Rockford, IL).

2.3. Antibodies

Antibodies used in this study were from the following sources: rabbit polyclonal anti-phospho p53 (Ser-15) (1:1000), rabbit monoclonal anti-SUMO-2/3 (1:1000) from Cell Signaling Technology (Danvers, MA); rabbit polyclonal anti-Cyclophilin B (1:5000) from Abcam Inc. (Cambridge, MA); rabbit polyclonal anti-SUMO-1 (1:1000) from Enzo Life Sciences (Farmingdale, NY) and all secondary antibodies (1:3000) were obtained from Jackson ImmunoResearch (West Grove, PA).

2.4. Cell injury models

(1) Cisplatin treatment of cells: HEK293 cells and RPTC were incubated respectively with 50 μM and 20 μM cisplatin in culture medium as previously [21,22]. To determine the effects of ROS inhibitors and p53 inhibitor, the inhibitors were added during cisplatin treatment. After incubation for an indicated time, the cells were analyzed by morphology for apoptosis or harvested with lysis buffer to collect cell lysates for various biochemical analyses. (2) ATP depletion: ATP depletion was induced in RPTC by azide treatment as before [20]. Briefly, RPTC were treated with 10 mM azide in glucose-free Krebs-Ringer bicarbonate buffer for time as indicated. After incubation, the cells were returned to full culture medium for recovery. After recovery for an indicated time, the whole cell lysates were collected for immunoblotting.

2.5. Animal model of AKI

C57BL/6 mice were originally purchased from the Jackson Laboratory and maintained in the animal facility of Charlie Norwood VA Medical Center at Augusta. The experimental protocols were approved by the Institutional Animal Care and User Committees of Charlie Norwood VA Medical Center.

2.5.1. Cisplatin AKI [21,23,24]

Male mice of 8 to 10 weeks were injected with a single dose of 30 mg/kg cisplatin by intraperitoneal injection to induce nephrotoxicity and kidneys were collected at 1 day, 2 day and 3 days after injection. Control animals were injected with saline.

2.5.2. Ischemic AKI [20,24,25]

Male mice of 8 to 10 weeks were anesthetized with one intraperitoneal injection of pentobarbital sodium (50–60 mg/kg). Flank incisions were made to expose renal pedicles for bilateral clamping for 30 min to induce renal ischemia. For ischemia only group, kidneys were harvested after 30 min of clamping. For reperfusion groups, clamps were released for reperfusion and after an indicated time, kidneys were harvested. For sham control, mice were conducted by an identical procedure but without renal pedicle clamping. Kidneys were collected after an indicated time for examination.

2.6. Analysis of apoptosis

Apoptosis in cell cultures was analyzed by morphological examination and caspase activity measurement.

2.6.1. Morphological examination of apoptosis

In brief, after various treatments, cells were stained with 10 μg/ml Hoechst 33342 and fixed with 4% paraformaldehyde. Then phase contrast and fluorescence microscopy were used to analyze cellular and nuclear morphologies. The typical apoptotic morphology is the formation of apoptotic bodies, nuclear condensation and fragmentation, and cellular shrinkage. The cells showing typical apoptotic morphology were counted to determine the percentage of apoptosis. For counting, each field has approximately 200 cells, and five fields were counted for each condition.

2.6.2. Caspase activity assay

Caspase activity was measured with an enzymatic assay by using the fluorogenic peptide carbobenzoxy-Asp-Glu-Val-Asp-7-amino-4 trifluoromethyl coumarin (DEVD.AFC), a substrate of executioner caspases including caspase-3, -6 and -7. In brief, cell lysates were collected in 1% Triton X-100 buffer and then added to (25 μg of protein/sample) an enzymatic reaction containing 50 μM DEVD.AFC for 60 min at 37 °C. The fluorescence signal of AFC liberated by caspase activity was measured with a GENios platereader (Tecan US Inc.) at an excitation of 360 nm and emission of 530 nm. A standard curve was constructed for each measurement by using free DEVD.AFC. The fluorescence reading of the enzymatic reaction was converted into the nanomolar amount of DEVD. AFC per mg protein per hour based on the standard curve is used as a measure of caspase activity.

2.7. Immunoblot analysis

Whole cell lysate and tissue lysate were collected in 2% SDS lysis buffer in the presence of 20 mM N-ethylamaleimide (NEM). Briefly, protein samples were resolved under reducing conditions on 4–12% gradient NuPAGE gels (Invitrogen, Carlsbad, CA), transferred to polyvinylidene difluoride (PVDF) membranes, blocked with 5% fat-free milk, and then probed with specific primary antibody overnight at 4 °C. After primary antibody incubation, the blots were washed and incubated with the horseradish peroxidase (HRP)-conjugated secondary antibody. Finally, the antigen specific signals were detected through incubation with the enhanced chemiluminescence (ECL) kit (Pierce, Rockford, IL). Cyclophilin B was used as loading control.

2.8. Statistical analysis

Qualitative data were expressed as means ± S.D. The statistical differences between two groups were determined by two-sample assuming equal variance *t*-test with Microsoft EXCEL 2007. $P < 0.05$ was considered to indicate significant differences.

3. Results

3.1. Changes in SUMOylation during renal ischemia–reperfusion

Our initial study investigated the SUMOylation change pattern in a mouse model of renal ischemia–reperfusion. To this end, male C57BL/6 mice of 8–10 weeks were subjected to bilateral clamping for 30 min to induce renal ischemia, followed by 0–48 h reperfusion. Kidney tissue lysate was collected at various time points for immunoblot analysis using specific antibodies to SUMO-1 or SUMO-2/3 to reveal SUMO-conjugated proteins. As shown in Fig. 1A, several SUMO-1-conjugated proteins were detected in control kidney tissues. Upon ischemia, there was a slight decrease in some of these protein bands. However, 8 h reperfusion led to a dramatic increase of SUMOylation, especially at

the high molecular weight range, resulting in a heavy smear. This increase disappeared during further reperfusion of 24–48 h (Fig. 1A). The changes in protein SUMOylation were confirmed by densitometric analysis of SUMOlyated protein smear of >72 kDa (Fig. 1B). For SUMO-2/3, there was a slight decrease during renal ischemia followed by a marked increase at 8 h reperfusion, and by 24–48 h, SUMOylation returned towards the basal control level (Fig. 1C, D). These results suggest a dynamic SUMOylation regulation during renal ischemia–reperfusion.

3.2. SUMOylation during ATP depletion and subsequent recovery in RPTC

We further examined protein SUMOylation in cultured renal tubular cells during ATP depletion and subsequent recovery, an in vitro model that partially recapitulates the in vivo condition of renal ischemia–reperfusion [26]. ATP depletion was induced by incubating RPTC in a glucose-free buffer containing azide, a mitochondrial respiration inhibitor at complex IV. After that, the cells were returned to normal culture medium for recovery. As shown in Fig. 2, compared with control cells, the levels of SUMOylated proteins by both SUMO-1 and SUMO-2/3 were suppressed markedly during 1–3 h of ATP depletion. Upon recovery, protein SUMOylation dramatically increased to levels that were higher than control (Fig. 2). The blotting result was verified by quantification via densitometry. The decrease of SUMOylation during ATP depletion is consistent with the ATP-dependence of SUMO conjugation. The induction of SUMOylation during early recovery of ATP-depleted cells echoes the finding of in vivo renal ischemia–reperfusion shown in Fig. 1.

3.3. Increase of protein SUMOylation in cisplatin AKI

Next, we determined whether SUMOylation occurs in cisplatin-induced nephrotoxic AKI. In the in vitro model, RPTC were incubated with 20 μM cisplatin. Following cisplatin incubation, an increase in global cellular SUMOylation by both SUMO-1 and SUMO-2/3 was detected (Fig. 3A, C). Similar results were obtained in cisplatin-treated HEK293 cells (Data not shown). In vivo, male C57BL/6 mice were intraperitoneally injected with a single dose of 30 mg/kg cisplatin to induce AKI. A massive rise in SUMO conjugations (both SUMO-1 and SUMO-2/3 conjugations) appearing as a smear at high molecular weight was detected at Day 2 and continued to Day 3 after cisplatin treatment (Fig. 3E, G). The induction of SUMOylation during cisplatin AKI was substantiated by densitometry analysis of the immunoblots (Fig. 3B, D, F, H). Together, these results demonstrate an induction of protein SUMOylation during cisplatin nephrotoxicity.

3.4. Effects of ROS inhibitors on SUMOylation induction during cisplatin treatment

To gain some insights into the mechanism of SUMOylation induction in AKI, we focused on cisplatin treatment. We first examined the involvement of ROS, because cisplatin-induced AKI is associated with oxidative stress [27,28], which has been shown to trigger SUMOylation. [29,30]. Specifically, we examined the effects of two antioxidants, DMTU and NAC, which were shown to attenuate oxidative stress and protect the cells against cisplatin-induced apoptosis in our previous work [31]. RTPC cells were incubated with 20 μM cisplatin in the absence or presence of 10 mM NAC or 10 mM DMTU for 8 h. As shown in Fig. 4, protein SUMOylation was significantly induced during cisplatin treatment (Fig. 4A and C: lane 2), but the induction was suppressed by DMTU (Fig. 4A and C: lane 3) or NAC (Fig. 4A and C: lane 4). We verified the results by densitometry analysis of the immunoblots from separate experiments (Fig. 4B, D). These results suggest the involvement of oxidative stress in SUMOylation induction in cisplatin AKI.

3.5. Effects of p53 inhibitor on SUMOylation induction during cisplatin treatment

p53 signaling pathway plays a significant role in cisplatin AKI [24, 32]. We wondered if SUMOylation induction by cisplatin is subjected to p53 regulation. To answer this question, we examined the effects of pifithrin-α, a specific pharmacologic inhibitor of p53. RTPC cells were incubated with 20 μM cisplatin in the absence or presence of 20 μM pifithrin-α for 8 h. We found that SUMO-1 conjugation was significantly induced during cisplatin treatment regardless the presence of pifithrin-α (Fig. 5A, B). However, the activation of SUMOylation by SUMO-2/3

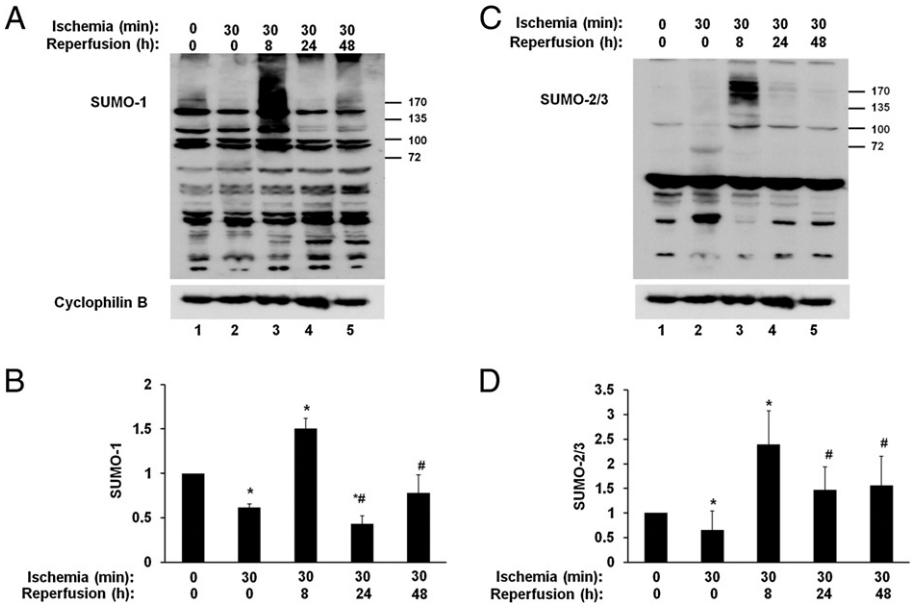

Fig. 1. Changes of SUMOylation during renal ischemia–reperfusion in mice. Male C57BL/6 mice were subjected to bilateral clamping for 30 min to induce renal ischemia followed by an indicated time of reperfusion. Sham control mice were subjected to an identical surgical procedure but without renal pedicle clamping. Renal tissues were analyzed by immunoblot analysis of SUMO-1 (A) and SUMO-2/3 (C). The blots were reprobed for Cyclophilin B as a loading control. (B) and (D): Densitometry analysis. Data are presented as mean ± SD; n ≥ 3. *P < 0.05, significantly different from untreated control. #P < 0.05, significantly different from the 8 h perfusion group.

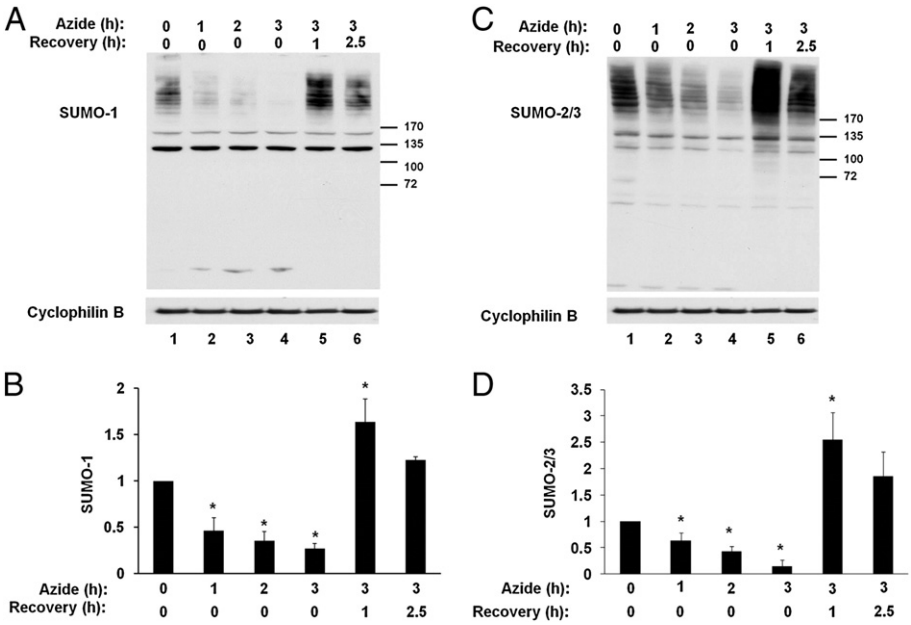

Fig. 2. Changes of protein SUMOylation during ATP depletion and recovery in RPTC. RPTC were subjected to 10 mM azide in glucose-free Krebs buffer for 0, 1, 2, 3 h. Then the cells of 3 h azide treatment were returned to full culture medium for 1 h or 2.5 h of recovery. Whole cell lysates were collected for immunoblot analysis of SUMO-1 (A) and SUMO-2/3 (C). The blots were reprobed for Cyclophilin B as loading control. (B) and (D): Densitometry analysis. Data are presented as mean ± SD; n ≥ 3. *P < 0.05, significantly different from untreated control.

was markedly suppressed by pifithrin-α (Fig. 5C, D). The inhibitory effect of pifithrin-α on p53 was verified by immunoblot analysis of phosphorylated p53 phosphorylation at serine-15 (Fig. 5A, B). These results indicate that p53 contributes to SUMOylation by SUMO-2/3, but not that by SUMO-1.

3.6. Inhibition of global protein SUMOylation sensitizes RPTC to apoptosis

There is evidence that SUMOylation induction during cell stress may act as a mechanism for cell survival [33–35]. The cell biologic function of SUMOylation during cisplatin treatment was unclear. To address this question, we tested the effect of ginkgolic acid (GA), a newly identified SUMOylation inhibitor that directly binds to E1 to inhibit the formation of the E1-SUMO intermediate resulting in the suppression of SUMOylation [36]. In pilot tests, we titrated the condition of GA treatment and found that 2 h pretreatment with 100 μM GA could effectively block SUMOylation induced by cisplatin without causing significant cytotoxicity (Fig. 6B, C). Compared with cisplatin-only group, 2 h pretreatment with GA suppressed cisplatin-induced SUMOylation by both SUMO-1 and SUMO-2/3 (Fig. 6A). Importantly, GA pretreatment

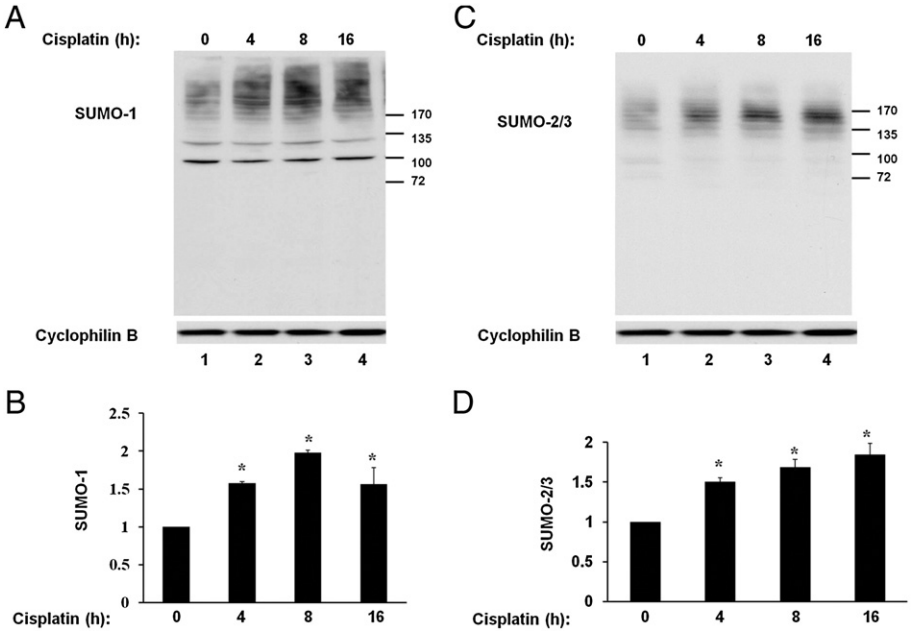

Fig. 3. . Protein SUMOylation in RPTC and kidneys during cisplatin treatment. RPTC were incubated with 20 μM cisplatin for 0 h, 4 h, 8 h, and 16 h. After incubation, whole cell lysates were collected for immunoblot analysis of SUMO-1 (A) and SUMO-2/3 (C). Male C57BL/6 mice were injected with 30 mg/kg cisplatin or saline as control. Renal tissues were collected at 0, 1, 2, or 3 days after cisplatin injection for immunoblot analysis of SUMO-1 (E) and SUMO-2/3 (G). The blots were reprobed for Cyclophilin B as loading control. (B), (D), (F) and (H): Densitometry analysis. Data are presented as mean ± SD; n ≥ 3. *P < 0.05, significantly different from untreated control.

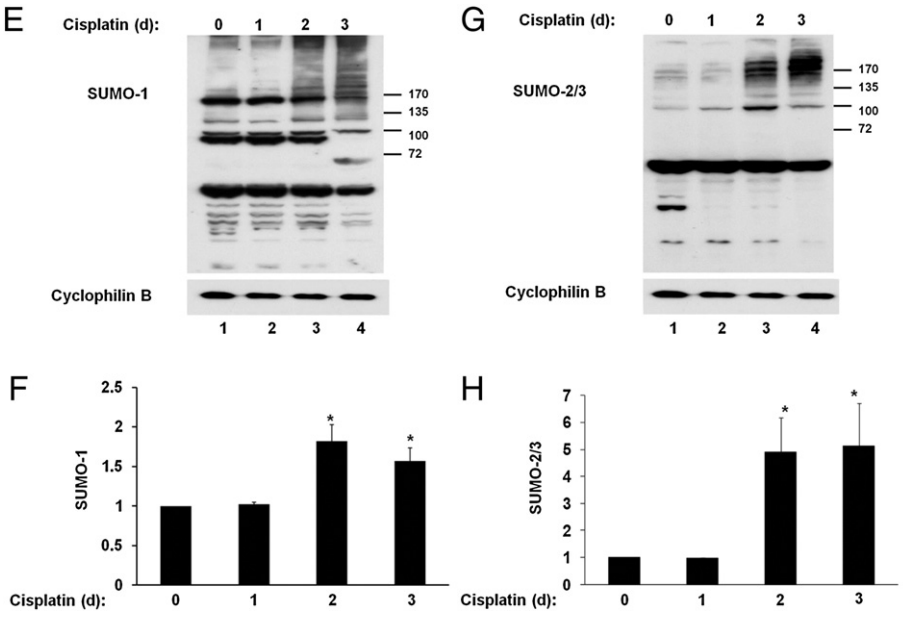

Fig. 3. (*continued*).

increased apoptosis during cisplatin treatment (Fig. 6B, C). The morphological analysis of apoptosis was confirmed by the measurement of caspase activity (Fig. 6D). Taken together, these data suggest that inhibition of SUMOylation sensitizes renal tubular cells to apoptosis, supporting a cytoprotective role of SUMOylation in cisplatin nephrotoxicity.

4. Discussion

During the last decade, SUMOylation has been recognized as an important post-translational modification that regulates protein stability, localization, interaction and activity. SUMOylation has also emerged as a major player in a broad array of cellular processes, including signal

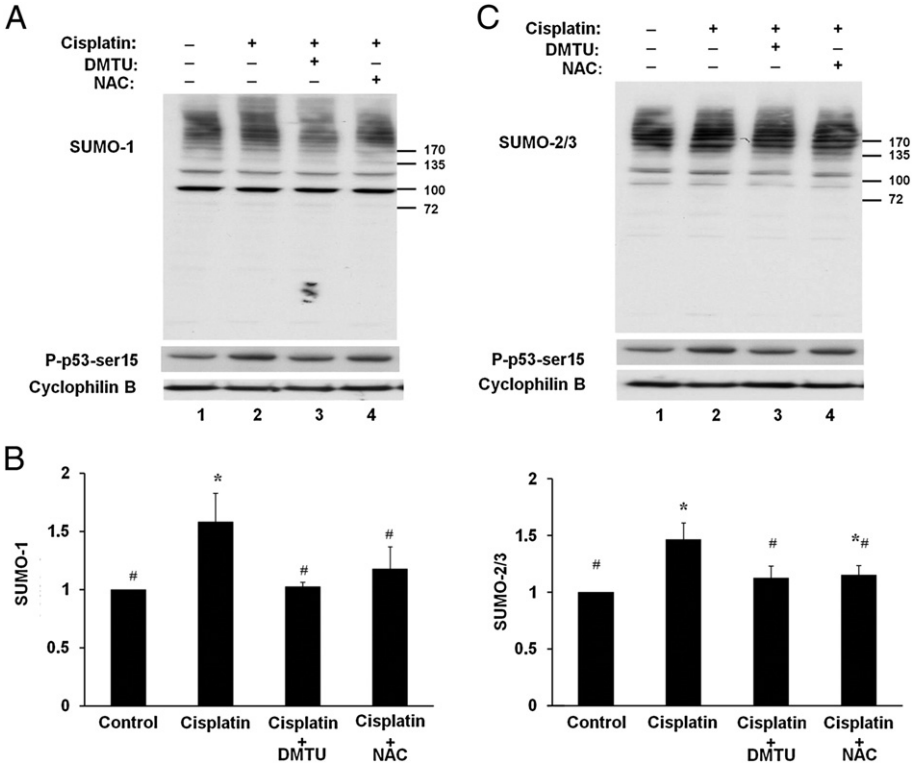

Fig. 4. Effects of NAC and DMTU on protein SUMOylation during cisplatin treatment in RPTC. RPTC were treated with 20 μM cisplatin in the absence or presence of 10 mM N-acetyl-cysteine (NAC) or 10 mM DMTU for 8 h. Whole cell lysate was collected for immunoblot analysis of SUMO-1 (A) and SUMO-2/3 (C). The blots were reprobed for Cyclophilin B as loading control. (B) and (D): Densitometry analysis. Data are presented as mean ± SD; $n \geq 3$. *$P < 0.05$, significantly different from untreated control; #$P < 0.05$, significantly different from the cisplatin-only group.

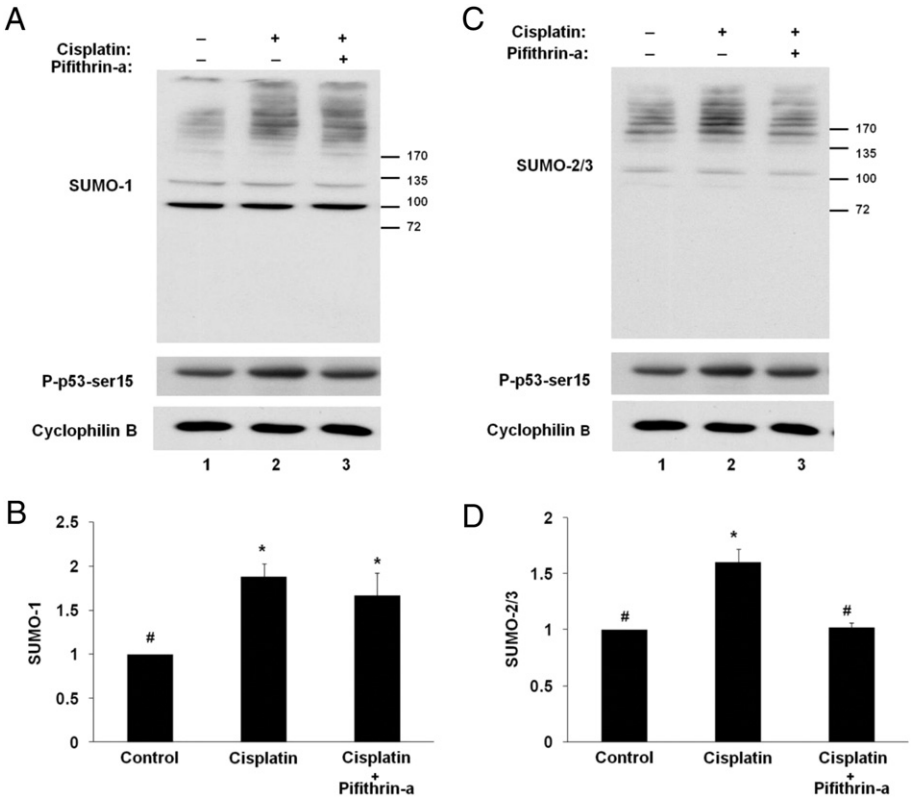

Fig. 5. Effects of pifithrin-α on protein SUMOylation during cisplatin treatment in RPTC. RTPC cells were treated with 20 μM cisplatin in the absence or presence of 20 μM pifithrin-α for 8 h. Whole cell lysate was collected for immunoblot analysis of SUMO-1 (A) and SUMO-2/3 (C). The blots were reprobed for Cyclophilin B as loading control. (B) and (D): Densitometry analysis. Data are presented as mean ± SD; n ≥ 3. *P < 0.05, significantly different from untreated control; #P < 0.05, significantly different from the cisplatin-only group.

transduction, nuclear transport, transcriptional regulation, maintenance of genome integrity and cell proliferation [37–39]. However, SUMOylation not only is an important regulator of normal cellular processes, but may also play a role in the pathogenesis of human diseases. Especially, SUMOylation is known to be activated by hypoxic, osmotic, oxidative, genotoxic and metabolic stresses [14–19]. More recent studies provide a biological link between the SUMOylation machinery and different human diseases, including cancer, heart failure, diabetes as well as neurodegenerative diseases [40–43]. However, very little is known about SUMOylation, its regulation, and pathophysiological role(s) in renal pathophysiology, especially in AKI.

In this study, changes in global protein SUMOylation were demonstrated in both ischemic and cisplatin nephrotoxic AKI. For ischemic AKI, we examined renal ischemia–reperfusion in mice and ATP depletion–recovery in RPTC. A decrease of SUMOylation was detected during renal ischemia and ATP-depletion (Figs. 1 and 2). This decrease was expected, because the enzymatic reaction of SUMOylation is ATP-dependent [11,12] and, renal ischemia in vivo and ATP-depletion in cultured tubular cells are known to result in a rapid cellular ATP deprivation [26,44]. Upon reperfusion or recovery, there was a marked increase or induction of SUMOylation (Figs. 1 and 2). This induction is consistent with the recent observation of increased SUMOylation in stroke models of the brain [45–47]. We have further demonstrated a time-dependent SUMOylation in kidney tissues and cells of cisplatin-induced AKI or nephrotoxicity. Together, these results provide the first evidence for a dynamic alteration of protein SUMOylation in AKI.

What are the underling mechanisms for the global changes in SUMOylation in AKI? In the present study, we focused on the cisplatin model to gain some initial clues. Our data suggest the involvement of oxidative stress (Fig. 4). Previous studies have demonstrated a complex relationship between oxidative stress and SUMOylation in mammalian cells. On one hand, severe oxidative stress was shown to increase SUMOylation, which may result from the inactivation of SUMO proteases by creation of an intra- or inter-molecular disulfide bridge [29,48]. On the other hand, low or moderate oxidative stress was shown to suppress global SUMOylation by introducing a disulfide bond between SUMO E1 and E2 enzymes at the catalytic cysteine residues or stabilizing SUMO proteases by formation of a disulfide bond in a regulatory element [17]. We specifically examined the effect of two antioxidants or ROS scavengers on SUMOylation during cisplatin treatment of RPTC (Fig. 4). Oxidative stress is associated with and contributes to cisplatin AKI [28,49–51]. Antioxidants protect renal tubular cells against cisplatin-induced apoptosis [31,52]. In this study, we showed that both antioxidants (NAC and DMTU) suppressed SUMOylation induction during cisplatin treatment (Fig. 4), supporting a role of oxidative stress. Furthermore, we observed that the change pattern of SUMOylation was correlated with p53 phosphorylation or activation (Fig. 4A and C). Notably, inhibition of p53 with pifithrin-α partially blocked SUMO-2/3 conjugation, but not SUMO-1-mediated SUMOylation (Fig. 5). This finding is intriguing and requires further in-depth investigation to understand the p53-mediated regulatory mechanism. Together, these observations indicate that the regulation of SUMOylation is very complex and involves multiple signaling pathways.

Functionally, upregulated SUMOylation has been implicated in cytoprotection for cell survival, at least under some stress conditions. For example, silencing SUMO-2/3 in primary cortical neurons increased cell death during transient oxygen/glucose deprivation [35]. Lee et al. further [33] demonstrated that focal cerebral ischemic damage is protected in Ubc9 transgenic mice through elevated global SUMOylation. Similarly, a more recent study found that enhanced SUMO-2/3 conjugation by down-regulating the deSUMOylation enzyme SENP3 in rat cortical neurons promoted cell survival after oxygen/glucose deprivation [34]. Our present data show that suppression of global SUMOylation by GA enhances apoptosis during cisplatin

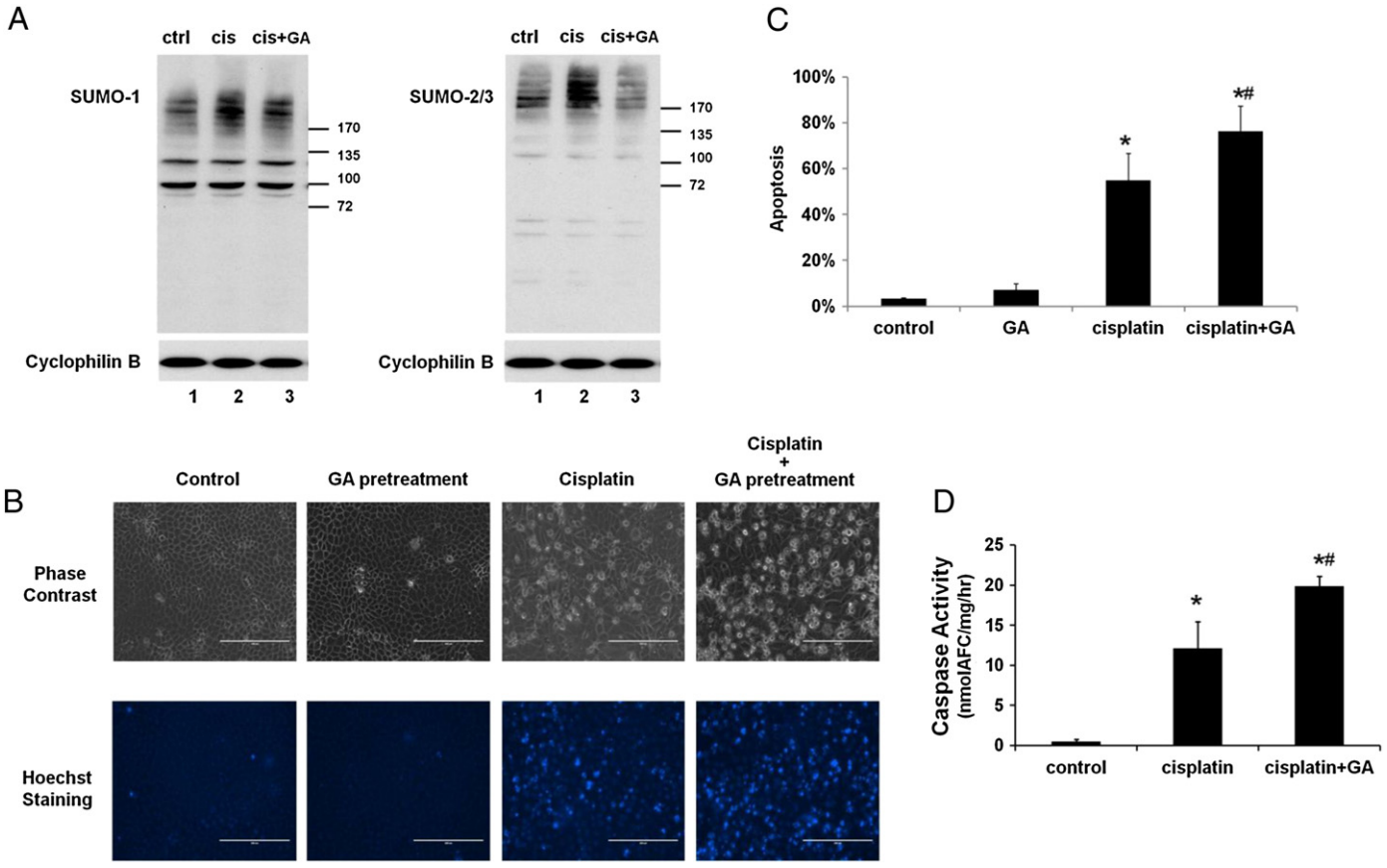

Fig. 6. Ginkgolic acid (GA) suppresses SUMOylation during cisplatin treatment and sensitizes cells to apoptosis. RTPC cells were treated with 20 μM cisplatin in the absence or presence of 2 h of 100 μM GA pretreatment. (A) Immunoblot. After 8 h cisplatin treatment, whole cell lysate was collected for immunoblot analysis of SUMO-1 and SUMO-2/3. (B) Cell and nuclear morphology. After 18 h cisplatin treatment, cells were stained with Hoechst 33342 and fixed with 4% paraformaldehyde. Then phase contrast and fluorescence microscopes were used to analyze cellular and nuclear morphology. (C) Percentage of apoptosis. The cells with apoptotic morphology were counted to determine the % of apoptosis. (D) Caspase activity. Cell lysate was collected to determine caspase activity by an enzymatic assay. Data in (C) and (D) are presented as mean ± SD; $n \geq 3$. *$P < 0.05$, significantly different from control; #$P < 0.05$, significantly different from the cisplatin-only group.

treatment of RPTC (Fig. 6), suggesting that SUMOylation plays a cytoprotective role in renal tubular cells.

There are several potential SUMOylated proteins that may be involved in AKI. For example, Drp1, the mitochondrial fission protein, contributes to cytochrome c release and apoptosis playing an important role in AKI [53]. SUMOylation of Drp1 impairs its localization to mitochondria and prevents mitochondrial fragmentation, cytochrome c release and apoptosis [34]. IκBα is another potential target of SUMOylation. IκBα is an inhibitor of NFκB and SUMOylation of IκBα by SUMO2/3 facilitates its disassociation from NFκB, which allows NFκB activation to result in a cell survival response [54]. HDAC2, a Class I deacetylase, can be modified by SUMO-1. SUMOylated HDAC2 deacetylates p53, which blocks the transcriptional activation of proapoptotic genes and attenuates DNA damage-induced apoptosis [55].

In summary, the present study has demonstrated the first evidence of SUMOylation during ischemic and cisplatin nephrotoxic AKI. Oxidative stress and p53 signaling may contribute to the induction of SUMOylation. Upon induction, SUMOylation may play a cytoprotective role against cell death and tissue damage in AKI.

Acknowledgements

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