Pakistan Veterinary Journal

ISSN: 0253-8318 (PRINT), 2074-7764 (ONLINE)
DOI: 10.29261/pakvetj/2025.339

RESEARCH ARTICLE

6-Gingerol Alleviates Acute Kidney Injury in Mice Via Activating Ampk/Nlrp3 Axis

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ARTICLE HISTORY (25-935)

Received: October 06, 2025
Revised: December 10, 2025
Accepted: December 24, 2025
Published online: December 26, 2025

Key words:
6-Gingerol
Acute kidney injury
Inflammasome
Inflammation
Ischemia-reperfusion

ABSTRACT

Acute kidney injury (AKI) commonly occurs following ischemia-reperfusion (I/R) during surgery or transplantation, and effective pharmacological treatments are limited. This study investigated the reno-protective effects and underlying mechanisms of 6-gingerol (6-G) in experimental mice of I/R-induced AKI. Renal I/R injury was induced in mice by bilateral renal pedicle clamping for 25 minutes followed by 24 hours of reperfusion. During reperfusion, mice received intragastric administration of 6-gingerol (50, 100 and 200mg/kg) every 8 hours. Renal function was evaluated by serum creatinine and blood urea nitrogen levels. Histopathological changes were assessed using hematoxylin and eosin and periodic acid–Schiff staining. Inflammatory responses were analyzed by measuring renal mRNA expression of interleukin-1β, interleukin-18, interleukin-6, and tumor necrosis factor-α. The expression of kidney injury markers (NGAL and KIM-1), inflammasome components (NLRP3, ASC & cleaved caspase-1) and phosphorylated/total AMP-activated protein kinase (AMPK) was examined using western blotting, immunohistochemistry, and immunofluorescence. 6-G treatment significantly improved renal function and attenuated tubular injury and glycogen deposition. It also markedly suppressed inflammatory cytokine expression and macrophage infiltration ($P<0.01$). Mechanistically, 6-G inhibited activation of the NLRP3/ASC/caspase-1 inflammasome while restoring AMPK phosphorylation. Notably, these protective effects were abolished by the AMPK inhibitor Compound C, indicating an AMPK-dependent mechanism. The findings demonstrated that 6-G alleviates renal dysfunction and tissue injury in I/R-induced AKI by activating AMPK and suppressing NLRP3 inflammasome-mediated inflammation, highlighting its therapeutic potential for AKI management.

To Cite This Article: Zhao Q, Yin X, Chang S, Che C, Wu T, Sun F and Chen Y, 2025. 6-Gingerol alleviates acute kidney injury in mice via activating ampk/nlrp3 axis. Pak Vet J, 45(4): 1980-1987. http://dx.doi.org/10.29261/pakvetj/2025.339

INTRODUCTION

Acute kidney injury (AKI) is a very common clinical syndrome, which is defined by a sudden loss of renal function (Ostermann *et al*., 2025). Although supportive care has improved, AKI remains strongly linked with both high morbidity rates and mortality; therefore, emphasizing the urgency of effective treatment (Pickkers *et al*., 2021). One of the most frequent causes of AKI is renal ischemia-reperfusion injury (IRI), which is especially prevalent in medical practice, particularly in major surgery, organ transplantation and septic shock (Rewa and Bagshaw 2014; Ronco *et al*., 2019). The pathophysiology of AKI caused by IRI is a complex and multifactorial phenomenon that includes oxidative stress, inflammatory cascades and programmed cell death pathways (Chouchani *et al*., 2014; Oh *et al*., 2023). It is worth noting that AKI is not just a clinically important disorder in human medicine, but it is also a vital issue in veterinary practice (Segev *et al*., 2024).

Renal IRI may be caused by trauma, surgery, dehydration, exposure to toxins or infectious diseases in veterinary patients. This results in a high level of morbidity and mortality similar to those of humans (Alaasam *et al*., 2024; Gryguc *et al*., 2024). It is worth mentioning that the veterinary research indicates that animal health requires efficient reno-protective measures (Tsunekawa and Sato, 2024). At the same time, this is also providing useful translational models to examine the pathogenesis of AKI

and the possibilities of therapeutic interventions. Therefore, advances in the understanding of the mechanisms of renal IRI are both significant in terms of the role they play in improving human clinical outcomes and contribute to the creation of specific therapies in veterinary medicine (Segev *et al*., 2024).

Substantial evidence underscores the central role of the NOD-like receptor pyrin domain-containing NLRP3 in modulating inflammatory responses throughout the development and progression of AKI (Anders, 2016; Tang *et al*., 2018; Lin *et al*., 2021). NLRP3 inflammasome drives caspase-1-dependent maturation and subsequent liberation of the pro-inflammatory cytokines, including interleukin-1β (IL-1β) and interleukin-18 (IL-18). Secreted cytokines, in turn, become major multipliers of renal inflammatory cascades, which worsen the tissue damage of kidneys (Lin *et al*., 2019; Wang *et al*., 2022). At the same time, any disruption of cellular energy metabolism is linked to the development of inflammasomes. AMPK has been shown to have reno-protective effects by alleviating oxidative stress, maintaining mitochondrial activity and inhibiting inflammatory signaling (Decleve *et al*., 2014). The role of AMPK in the protection of AKI has also been reinforced in recent research, showing that pharmacological stimulation of AMPK suppresses the function of the NLRP3 inflammasome directly, thereby preventing the development of renal injury in the face of ischemic injuries (El-Maadawy *et al*., 2022; Zhu *et al*., 2025).

Plant-derived natural compounds have become the target of therapeutic candidates because of their pleiotropic biological action and excellent safety profiles in most cases (Kumar *et al*., 2021; Rodrigues, 2024). These natural compounds include but are not limited to 6-Gingerol (6-G), the main bioactive constituent of ginger (Zingiber officinale), which is commonly recognized to have a strong antioxidant, anti-inflammatory and anti-apoptotic properties (Angelopoulou *et al*., 2022; Wu *et al*., 2022; Ghosh *et al*., 2023). Coordinated data support the idea that the use of 6-G has protective effects in various disease models, such as cardiovascular diseases (Ma *et al*., 2021) and hepatic injury (Yu *et al*., 2024) which prove the scope of pharmacological opportunities. However, its effectiveness in AKI has not been well studied, and the finer details of how its mechanisms the reno-protective effects are not well understood.

Considering the growing awareness of the crosstalk between energy metabolism and inflammatory signaling, and the regulatory relationship between AMPK activation and NLRP3 inflammasome inhibition, in particular (Pu *et al*., 2021; Cai *et al*., 2025). it is reasonable to hypothesize that 6-G may attenuate renal injury though AMPK/NLRP3 pathway. Elucidating this mechanism would not only expand our understanding of the pharmacological actions of 6-G but also provide novel insights into its potential as a therapeutic candidate for ischemic AKI. Thus, the present was planned to investigate the protective effects of 6-G against rodent AKI induced by IRI. The aim was to further explore whether its renoprotections are mediated through AMPK/NLRP3 cascades.

MATERIALS AND METHODS

Study design and experimental animals: Male C57BL/6 mice (9 weeks, 22-25g; Beijing Huafukang Biotechnology, China) were raised in a standard environment. For the primary experiment, mice were randomized into following five groups, each group having 10 mice (Table 1). I/R was induced by clamping both renal pedicles for 25 minutes under isoflurane anesthesia, followed by reperfusion as described by Cao *et al*. (2020). Sham mice underwent identical surgery without vascular clamping. 6-G was administered intra-gastrically (50, 100 or 200mg/kg, every 8 hours) after reperfusion (Ajayi *et al*., 2015). Mice were sacrificed after 24 hours, and blood samples were obtained via the facial vein. All animal experiments were approved by the Institutional Animal Care and Use Committee of the China Medical University (CMUKT2025188). To verify AMPK involvement, an additional set of mice was assigned to five groups: Sham, I/R, I/R+Compound C (CC, 10mg/kg), I/R+6-G-H (200mg/kg), and I/R+6-G-H+CC.

Table 1: Grouping of experimental animals along with dosage regime used for experiment

Group No.	Grouping
A	Sham group (control group)
B	I/R group (model group)
C	I/R+6-G-L group (low-dose treatment group, 50mg/kg 6-G)
D	I/R+6-G-M group (medium-dose treatment group, 100mg/kg 6-G)
E	I/R+6-G-H group (high-dose treatment group, 200mg/kg 6-G)

Renal function and histology: Serum creatinine (SCr, CB10746-Mu, COIBO BIO, Shanghai, China) and blood urea nitrogen (BUN, No. CB10533-Mu, COIBO BIO, Shanghai, China) were measured with commercial kits (Cao *et al*., 2020). Furthermore, urine output was recorded, and kidney-to-body weight ratio (KW/BW) was also calculated (Han *et al*., 2021). Then, the kidneys were exposed to H&E and PAS staining.

Immunohistochemistry analysis: Kidney sections (4μm) were processed for antigen retrieval and incubated with anti-neutrophil gelatinase-associated lipocalin (NGAL) (No. ab125075, Abcam, USA), anti-kidney injury molecule-1 (KIM-1) (No. ab316854, Abcam, USA), or anti-NLRP3 (No. ab270449, Abcam, USA). The remaining steps are all standardized as described by Wang *et al*. (2022).

Immunofluorescence analysis: Sections were antigen-retrieved, blocked and incubated with F4/80, NLRP3 or ASC antibodies, followed by fluorescent secondary antibodies and DAPI nuclear staining. Images were acquired by confocal microscopy (Wang *et al*., 2022).

Real-Time PCR assay: Real time PCR was used to determine the mRNA expression of IL-1β, IL-6, IL-18 and TNF-α and the experimental procedures were all been standardized (Lin *et al*., 2019). Primer sequences were in supplementary materials.

Western blotting assay: Renal tissue proteins were extracted, quantified, separated by SDS-PAGE and transferred to PVDF membranes and target proteins NGAL (No. ab125075, Abcam, USA), KIM-1 (No. ab316854, Abcam, USA), NLRP3 No. ab270449, Abcam, USA), ASC (No. ab309497, Abcam, USA), cleaved caspase-1 (No. ab207802, Abcam, USA), AMPK (No. ab23047, Abcam, USA), p-AMPK (No. ab23875, Abcam, USA), IL-1β (No. ab216995, Abcam, USA), IL-18 (No. ab207323, Abcam,

USA)) were detected with specific antibodies and quantified via ImageJ, using GAPDH as control (Lin *et al*., 2019).

Statistical analysis: Data were analyzed and given as mean ± standard deviation. One-way ANOVA followed by Tukey's test was applied with $P<0.05$ considered significant and GraphPad Prism was used to draw statistical graphs.

RESULTS

Renal function and AKI in mice: The renal I/R group (model group) exhibited increased SCr (Fig. 1A) and increased BUN (Fig. 1B) compared with the sham group (control group) ($P<0.01$); these elevations were dose-dependently reversed by 6-G ($P<0.01$). Additionally, the I/R group showed higher KW/BW ratio (Fig. 1C) and lower urine output (Fig. 1D) relative to the sham group ($P<0.01$), with these changes significantly attenuated by 6-G treatment ($P<0.01$). At the molecular level, immunohistochemistry revealed robust induction of NGAL and KIM-1 in I/R kidneys (Fig. 1E), which was markedly suppressed by 6-G in a dose-dependent manner (Fig. 1E). Western blot analysis further confirmed the upregulation of NGAL and KIM-1 protein expression following I/R and their significant reduction after 6-G treatment ($P<0.01$; Fig. 1F). Together, these results demonstrate that 6-G markedly improves renal function, mitigates structural injury, and suppresses the expression of key renal injury biomarkers in I/R-induced acute kidney injury.

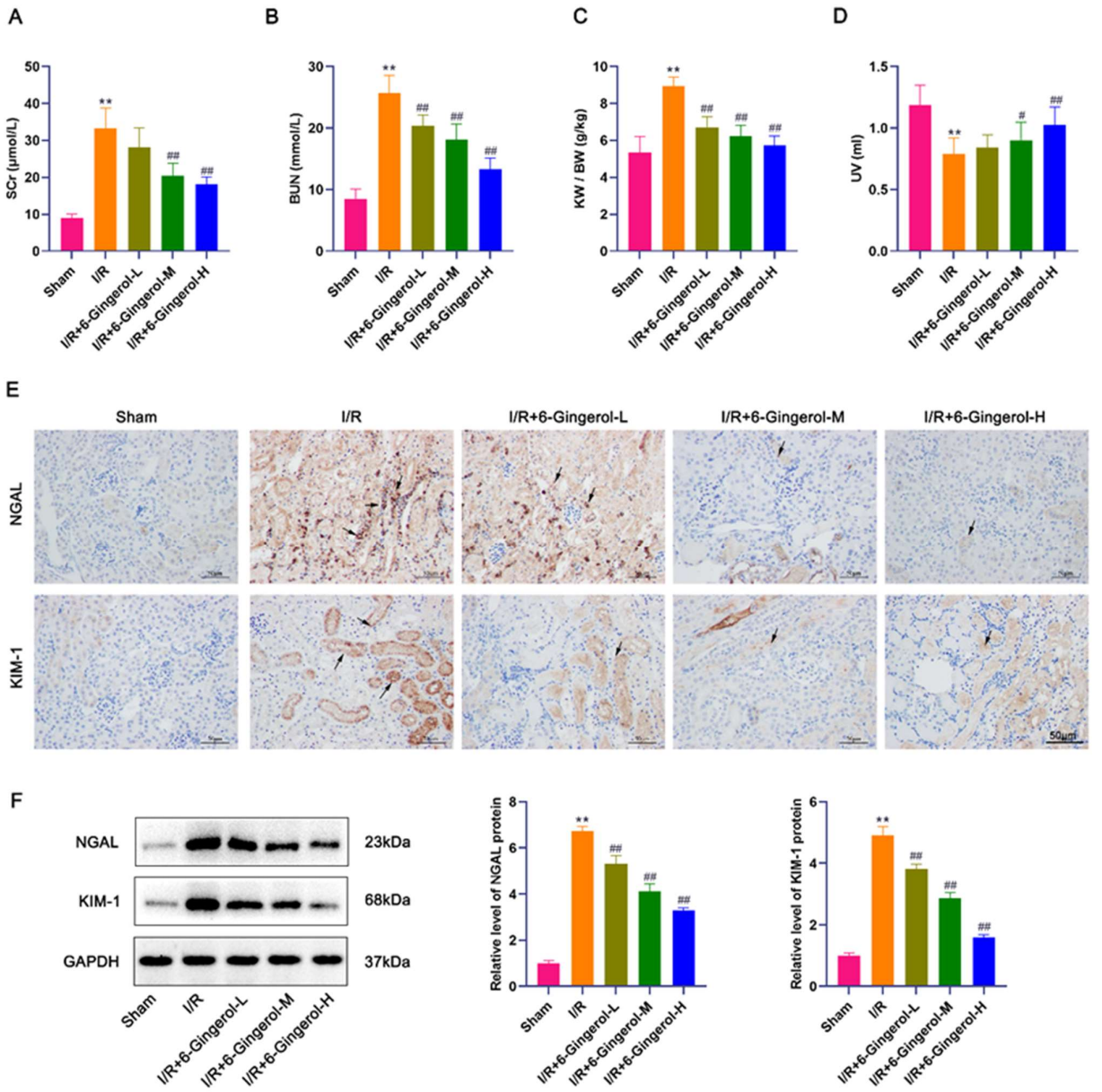

Fig. 1: 6-G Improves Renal Function and Attenuates AKI in mice: (A, B) Serum creatinine (SCr) and blood urea nitrogen (BUN) were elevated after I/R and reduced by 6-G in a dose-dependent manner. (C, D) Kidney weight-to-body weight ratio (KW/BW) increased and urine output decreased after I/R; both were ameliorated by 6-G. (E, F) Immunohistochemistry showed marked induction of NGAL and KIM-1 in I/R kidneys, which was suppressed by 6-G. (G) Western blot confirmed increased NGAL and KIM-1 expression after I/R and their reduction following 6-G treatment. Data are shown as mean ± SD (n=6-8). **P<0.01 vs. sham; ##P<0.01 vs. I/R (one-way ANOVA).

Renal histopathological alterations induced by I/R: Histological observations further validated the protective effect of 6-G against I/R-induced renal injury HE staining revealed severe structural damage in the I/R group, characterized by glomerular swelling, tubular dilation, and loss of brush borders compared with the sham group (Fig. 2A). Treatment with 6-G markedly attenuated these pathological changes in a dose-dependent manner. Both at 100 and 200mg/kg 6-G treatment showed a significant restoration of normal morphology (Fig. 2A). PAS staining further demonstrated prominent glycogen deposition and tubular injury in I/R group (model group), which were substantially alleviated by 6-G administration (Fig. 2B). Together, these findings indicate that 6-G effectively mitigates I/R-induced histopathological damage in renal tissues.

Renal inflammation after AKI: To investigate the anti-inflammatory effects of 6-G, inflammatory cytokine expression in renal cortex tissues was examined. Compared with the sham group (control group), I/R markedly increased mRNA levels of IL-1β, IL-18, IL-6 and TNF-α ($P<0.01$; Fig. 3A-D) and Administration of 6-G significantly suppressed these elevations ($P<0.01$; Fig. 3A-D). In addition, immunofluorescence staining of F4/80 revealed robust macrophage infiltration in model I/R kidneys, which was markedly alleviated by 6-G treatment (Fig. 3E). Collectively, these findings illustrate that 6-G exerts robust anti-inflammatory effects in I/R-induced AKI by downregulating pro-inflammatory cytokines and attenuating macrophage accumulation.

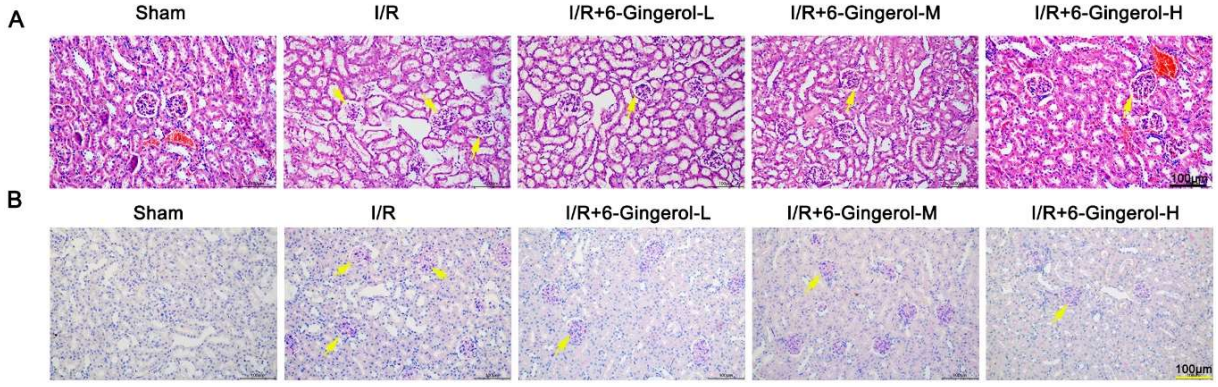

Fig. 2: 6-G alleviates renal histopathological damage after I/R: (A) HE staining showed glomerular swelling, tubular dilation, and loss of brush borders in I/R kidneys, which were ameliorated by 6-G in a dose-dependent manner. (B) PAS staining revealed glycogen deposition and tubular injury in I/R kidneys, both of which were markedly reduced after 6-G treatment. Data are shown as mean±SD (n=6-8). **P<0.01 vs. sham; ##P<0.01 vs. I/R (one-way ANOVA).

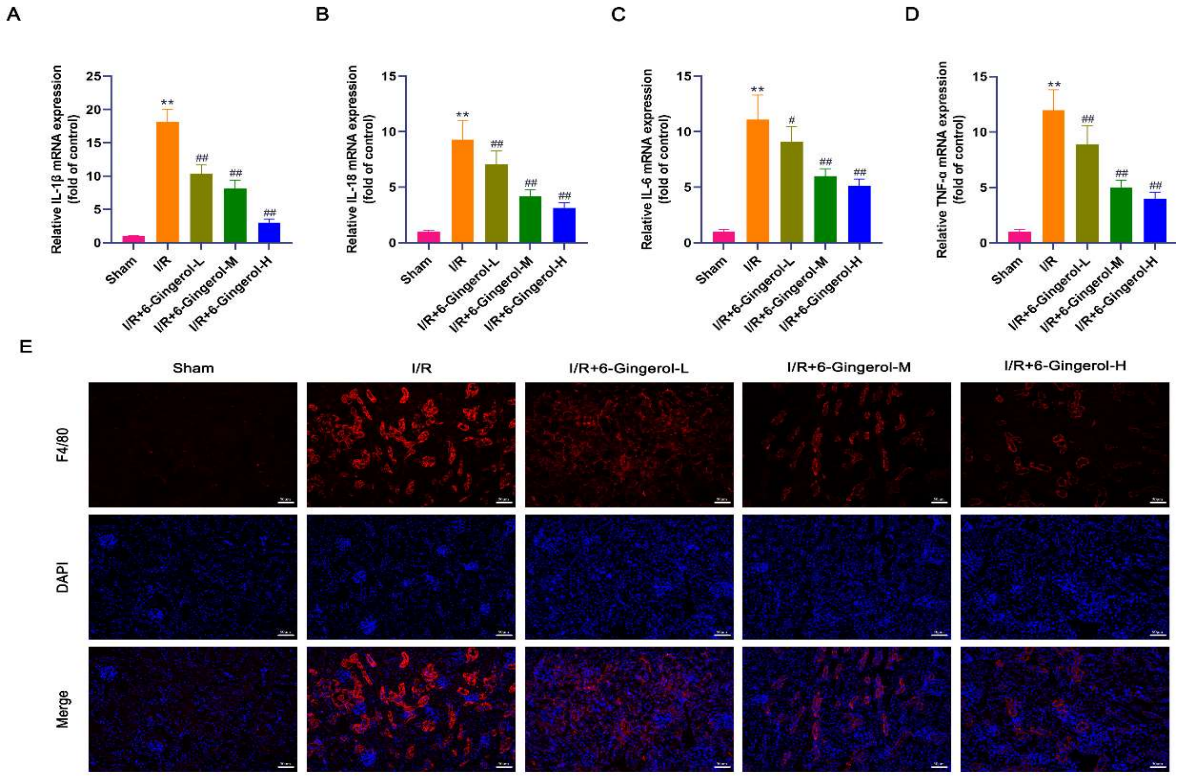

Fig. 3: 6-G Attenuates renal inflammation after AKI: (A-D) qRT-PCR analysis showed increased IL-1β, IL-18, IL-6, and TNF-α expression in I/R kidneys, which were significantly reduced by 6-G in a dose-dependent manner. (E) Immunofluorescence staining of F4/80 revealed enhanced macrophage infiltration after I/R, which was markedly attenuated by 6-G treatment. Data are shown as mean±SD (n=6-8). **P<0.01 vs. sham; ##P<0.01 vs. I/R (one-way ANOVA).

Inhibition of activation of the NLRP3/ASC/Caspase-1 pathway: Western blot analysis showed elevated protein levels of NLRP3, ASC, cleaved caspase-1, IL-1β and IL-18 in the I/R group (model group) relative to Sham group (control group), while 6-G treatment dose-dependently suppressed the expression of these inflammasome components (P<0.01; Fig. 4A). Consistently, immunofluorescence staining revealed increased NLRP3 fluorescence intensity in I/R-injured kidneys, which was notably reduced after 6-G administration (Fig. 4B). Together, this data indicate that 6-G mitigates I/R-induced AKI, at least partially, by inhibiting activation of the NLRP3 inflammasome.

Inhibition of NLRP3 inflammasome activation: Western blot analysis showed that renal I/R markedly suppressed AMPK phosphorylation compared with Sham group (control group) (P<0.01; Fig. 5A), which was reversed by 6-G treatment (P<0.01; Fig. 5A). Western blot results demonstrated that CC administration abolished the inhibitory effect of 6-G on NLRP3 inflammasome activation (P<0.01; Fig. 5B). Consistently, qRT-PCR analysis revealed that 6-G markedly reduced the production of inflammatory factors (P<0.01; Fig. 5C-F). These findings indicate that the protective effects of 6-G against I/R-induced AKI are largely mediated through activation of the AMPK/NLRP3 signaling axis.

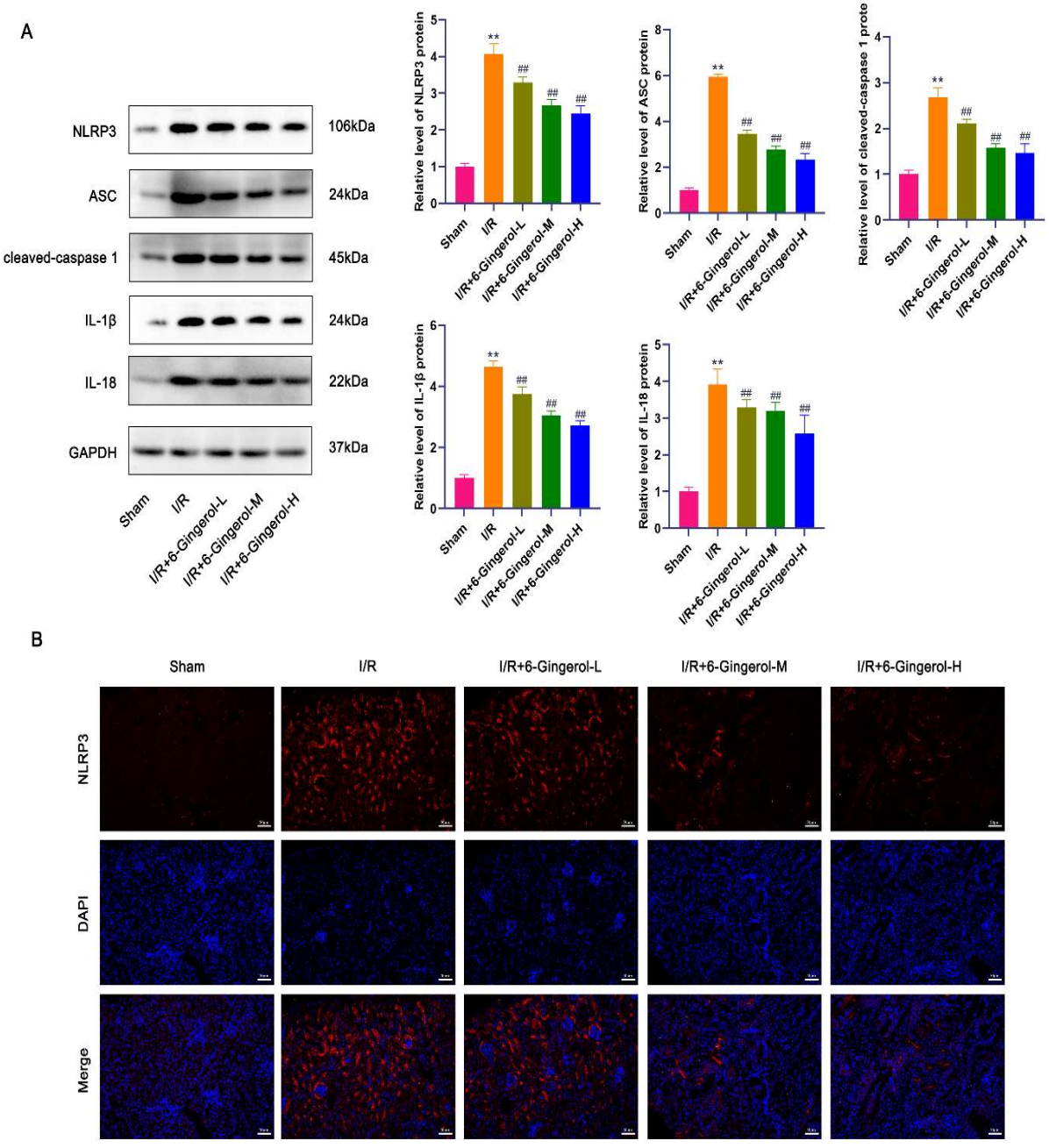

Fig. 4: 6-G inhibits activation of the NLRP3/ASC/Caspase-1 pathway: (A) Western blot analysis showed upregulation of NLRP3, ASC, cleaved caspase-1, IL-1β, and IL-18 in I/R kidneys, which were significantly reduced by 6-G in a dose-dependent manner. (B) Immunofluorescence staining revealed increased NLRP3 expression after I/R, which was markedly attenuated by 6-G treatment. Data are presented as mean ± SD (n=6-8). **P<0.01 vs. sham; ##P<0.01 vs. I/R (one-way ANOVA).

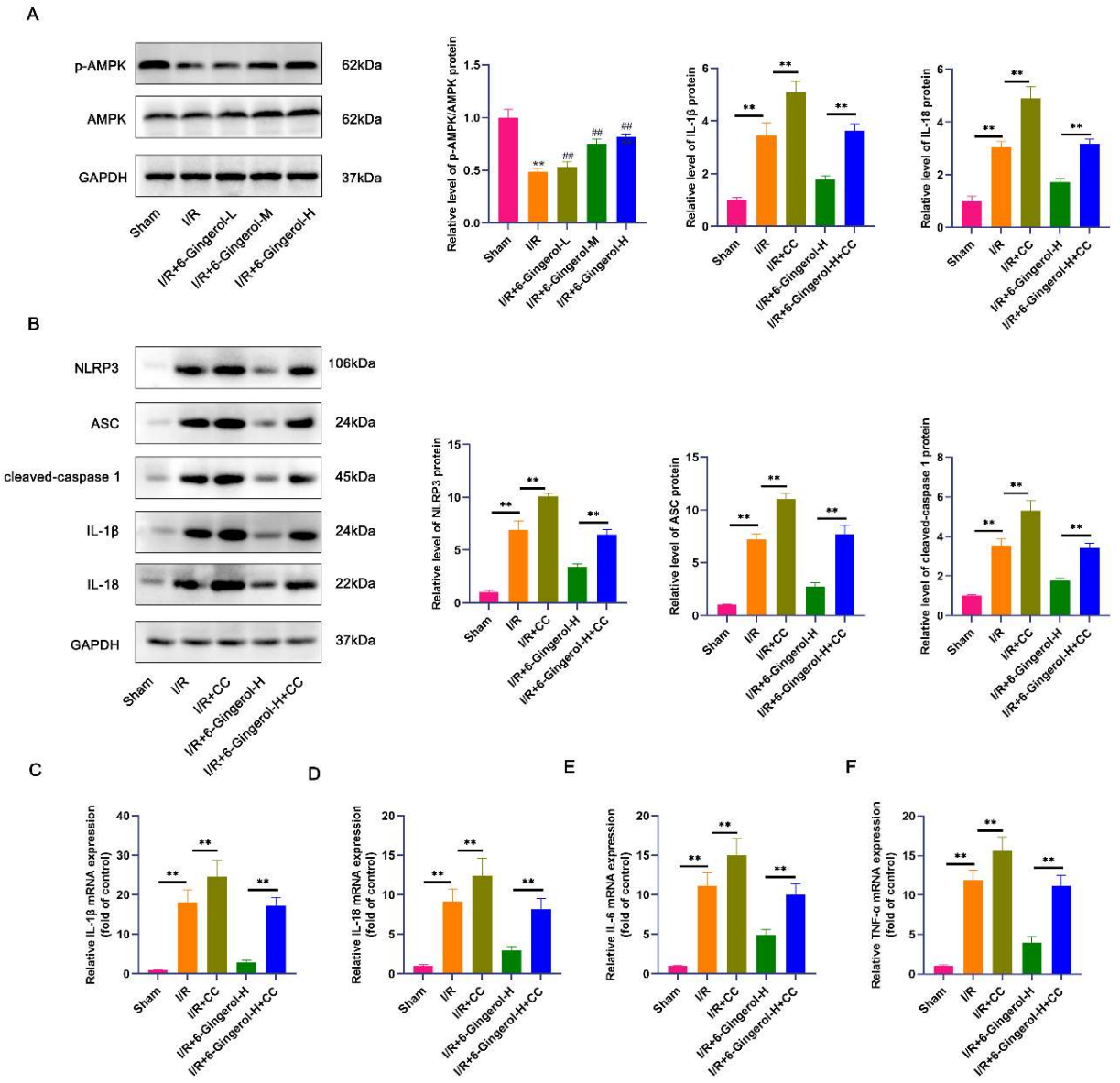

Fig. 5: 6-G Inhibits NLRP3 inflammasome activation through an AMPK-dependent mechanism: (A) Western blot analysis showed decreased p-AMPK after I/R, which was restored by 6-G in a dose-dependent manner. (B) Inhibition of AMPK by Compound C abolished the suppressive effects of 6-G on NLRP3, ASC, cleaved caspase-1, IL-1β, and IL-18 expression. (C-F) qRT-PCR analysis demonstrated reduced IL-1β, IL-18, IL-6, and TNF-α mRNA levels after 6-G treatment, which were reversed by Compound C. Data are presented as mean±SD (n=6-8). **P<0.01 vs. the indicated group.

DISCUSSION

Acute kidney injury is a significant cause of morbidity and mortality in both clinical and veterinary practice. It often occurs because of I/R injury during surgeries, organ transplantation or trauma (Messerer *et al.,* 2021). In veterinary medicine, AKI is often seen in companion animals like dogs and cats as well as in livestock (Segev *et al.,* 2024). Despite the progress in veterinary medicine, mortality rates of AKI in animals remain unacceptably high because of the deficiency of pharmacological treatments currently in use (Keir and Kellum, 2015; Segev *et al.,* 2024). The pathophysiological mechanisms that underlie AKI are complex and multifactorial and include oxidative stress, activation of inflammatory cascades, mitochondrial dysfunction and regulated cell death pathways (Wang and Zhang, 2022). Among these, AMPK signaling pathway has emerged as a critical cellular energy balance and stress adaptation regulator. Meanwhile, the NLRP3 inflammasome is central to mediating sterile inflammation in renal injury (El-Maadawy *et al.,* 2022; Jie *et al.,* 2022). Aberrant activation of the inflammasome complex, including the NLRP3 inflammasome, especially the NLRP3/ASC/caspase-1 signaling pathway, causes excessive release of pro-inflammatory cytokines causing further renal damage (Islamuddin and Qin, 2024).

In the present study, it was demonstrated that 6-G in ginger extract which is bioactive against I/R induced AKI with detected enhancing renal function, reduction of histological lesions, decreased inflammatory infiltration and suppression of inflammasome activations including the NLRP3 by depending on the AMPK pathway. Our biochemical results revealed that I/R injury causes a significant renal dysfunction as shown by the elevation in

SCr and BUN levels, renal edema and decreased urine output. These observations are consistent with the classical features of AKI pathophysiology (Gaut and Liapis, 2021; Pickkers *et al.,* 2021). However, the administration of 6-G was able to ameliorate these biochemical and functional abnormalities in a dose-dependent way. This protective effect of 6-G is in line with the reports on other natural phytochemicals, such as resveratrol, curcumin and quercetin, which have been shown to have similar reno-protective effects in I/R-induced AKI. These compounds have their beneficial effects by reducing oxidative stress and improving mitochondrial integrity (Kaur *et al.,* 2016; Bienholz *et al.,* 2017; Huang *et al.,* 2022).

Our experiment contributes to this body of evidence by discovering 6-G as another promising bioactive compound that could be used to restore the renal functions, thus the therapeutic efficacy of plant-derived compounds to the treatment of AKI. Biochemical analysis was also done with histological assessment that established that I/R injury resulted in severe necrosis of the tubular epithelial cell, the development of luminal cast and glycogen accumulation which are characteristic of renal injury. These structural abnormalities however were greatly ameliorated by the treatment with 6-G. We have corroborated the current literature that demonstrates the use of ginger extract in the prevention of structural injury in hepatic and myocardial I/R models using its antioxidative and anti-apoptotic properties (Sabina *et al.,* 2011; Yu *et al.*, 2024; Yang *et al.,* 2025). Also, other bioactive agents like ginsenosides and berberine which have demonstrated potential in the management of kidney injury due to their impact on several pathological mechanisms have been described to exert similar reno-protective effects (Chang *et al.,* 2023; Yang *et al.,* 2024). The implications of these studies are that phytochemicals could be used as multi-purpose therapeutic reagents capable of treating an array of various mechanisms of AKI, with the roots of oxidative damage to inflammation.

One of the major processes in the pathogenesis of post-IR renal injury is inflammation. We have observed large rises in the concentrations of key pro-inflammatory cytokines such as IL-1 beta, IL-18, IL-6 and TNF-alpha and the presence of macrophages in renal tissue in our study. These inflammatory modifications were greatly reversed by the aid of the treatment with 6-G and it is also supporting 6-G as an anti-inflammatory agent. The given result is in line with the earlier research that has shown the capacity of 6-G to suppress the NF-kB and the MAPK signaling in neuroinflammation models, arthritis and metabolic disease (Abusarah *et al.,* 2017; Song *et al.,* 2019; Angelopoulou *et al.,* 2022). Other established agents to exacerbate the harm to kidneys and slow the recovery of tissue in response to AKI are the recruitment of macrophages and cytokines (Cliff *et al.,* 2024). Blockage of such pathways of inflammation has been demonstrated to deepen the course AKI and enhance the renal outcome (Chang *et al.,* 2023; Yang *et al.*, 2024; Hu *et al.,* 2025). Therefore, our findings are highly indicative that the anti-inflammatory effect of 6-G has a significant role in its reno-protective effects.

Moreover, our paper gives direct information on the fact that AMPK/NLRP3 inflammasome axis plays a role in the protective effect of 6-G in AKI caused by I/R. AMPK is a supreme controller of cellular energy homeostasis and stress response, and the activation of it was found to possess a protective role against numerous forms of cellular damage, such as renal ischemia (Luo *et al.*, 2023). Here we discovered that the 6-G supplementation was effective to induce the action of the protein kinase AMPK and the outcome was the inactivation of the functioning of the type 11 inflammasome which consequently reduced the synthesis of the pro-inflammatory cytokines. One primary mediator of sterile inflammations involved in improving kidney injury in I/R is the NLRP3 inflammasome (El-Maadawy *et al.,* 2022). The phosphorylase enzyme and NLRP3 modulation with 6-G has more than a two-fold-pronged effect on the impact of I/R-induced renal damages and potential major therapeutic benefits, both preclinical and clinical. Although the current study has some promising results, the study has numerous limitations that should be overcome in the future research.

First, the research examined was largely based on short-term outcomes (24 hours after reperfusion), and further research on the long-term outcomes of 6-G on renal repair, fibrosis and survival are justified. Besides, pharmacokinetics of 6-G like its absorption, distribution, metabolism and excretion has not been explored completely. More research is required to streamline the dosage and route of 6-G, especially in large animals, to be able to translate the possibility to clinical and veterinary practice. Furthermore, there is need to determine the safety profile of the use of 6-G in the long run and any possible side effects or even toxicity expected. Lastly, our finding indicates that 6-G offers significant defense against I/R-induced AKI as indicated by the 6-G capacity to recover renal function, decrease histopathological damages, and balance the inflammatory and cellular stressor systems. These discoveries render 6-G an excellent prospect of future development as a therapeutic agent of AKI that may find application in the clinical and veterinary field. Nevertheless, research work is still necessary to comprehend the long-term outcomes and the way to optimize the dosage and explore the pharmacokinetic characteristics to make 6-G applicable to real life treatments to AKI in both human beings and animals.

Conclusions: Acute kidney injury is a commonly occurring condition reported during surgery or transplantation with limited pharmacological treatments. This study demonstrated that 6-G confers significant protection against I/R-induced AKI by improving renal function, reducing tubular, inflammatory injury and inhibiting NLRP3 inflammasome activation through AMPK-dependent signaling. These findings not only highlight 6-G as a promising candidate for ischemic AKI therapy but also underscore its translational potential in both human and veterinary medicine. Future work should explore its long-term efficacy, mechanistic complexity, and clinical feasibility.

Authors contribution: Qian Zhao and Xiaoming Yin: designed and performed most of the experimental work, analyzed the data, and drafted the initial manuscript. Shuang Chang and Chengyi Che: Assisted in animal model establishment. Tengfei Wu and Feifei Sun: Conceived and designed the research, provided critical guidance on experimental design. Yixin Chen: Supervised the study, data interpretation, revised the manuscript, and finalized the manuscript for submission.

Declaration of competing interest: No potential conflict of interest was reported by the author(s).

REFERENCES

Ajayi BO, Adedara IA and Farombi EO, 2015. Pharmacological activity of 6-gingerol in dextran sulphate sodium-induced ulcerative colitis in BALB/c mice. Phytother Res 29:566-572.

Alaasam ER, Janabi AM, Al-Buthabhak KM *et al*., 2024. Nephroprotective role of resveratrol in renal ischemia-reperfusion injury: a preclinical study in Sprague-Dawley rats. BMC Pharmacol Toxicol 25:82.

Anders HJ, 2016. Of Inflammasomes and Alarmins: IL-1β and IL-1α in kidney disease. J Am Soc Nephrol 27:2564-2575.

Angelopoulou E, Paudel YN, Papageorgiou SG *et al*., 2022. Elucidating the beneficial effects of Ginger (*Zingiber officinale* Roscoe) in Parkinson's Disease. ACS Pharmacol Transl Sci 5:838-848.

Cai Z, Sun F, Wang Q *et al*., 2025. Icariin alleviates cardiomyocyte pyroptosis through AMPK-NLRP3 pathway to ameliorates diabetic cardiomyopathy. Int Immunopharmacol 156:114690.

Cao JY, Zhou LT, Li ZL *et al*., 2020. Dopamine D1 receptor agonist A68930 attenuates acute kidney injury by inhibiting NLRP3 inflammasome activation. J Pharmacol Sci 143:226-233.

Chouchani ET, Pell VR, Gaude E *et al*., 2014. Ischaemic accumulation of succinate controls reperfusion injury through mitochondrial ROS. Nature 515:431-435.

Declèves AE, Sharma K and Satriano J, 2014. Beneficial effects of AMP-Activated protein kinase agonists in kidney ischemia-reperfusion: autophagy and cellular stress markers. Nephron Exp Nephrol 128:98-110.

El-Maadawy WH, Hassan M, Abdou RM *et al*., 2022. 6-Paradol alleviates Diclofenac-induced acute kidney injury via autophagy enhancement-mediated by AMPK/AKT/mTOR and NLRP3 inflammasome pathways. Environ Toxicol Pharmacol 91:103817.

Ghosh S, Das B, Haldar PK *et al*., 2023. 6-Gingerol contents of several ginger varieties of Northeast India and correlation of their antioxidant activity in respect to phenolics and flavonoids contents. Phytochem Anal 34:259-268.

Gryguc A, Maciulaitis J, Mickevicius L *et al*., 2024. Prevention of transition from acute kidney injury to chronic kidney disease using clinical-grade perinatal stem cells in non-clinical study. Int J Mol Sci 25:9647.

Han BH, Lee HK, Jang SH *et al*., 2021. Effect of Geumgwe-Sinkihwan on renal dysfunction in ischemia/reperfusion-induced acute renal failure mice. Nutrients 13:3859.

Kumar S, Mittal A, Babu D *et al*., 2021. Herbal medicines for diabetes management and its secondary complications. Curr Diabetes Rev 17:437-456.

Lin Q, Li S, Jiang N *et al*., 2021. Inhibiting NLRP3 inflammasome attenuates apoptosis in contrast-induced acute kidney injury through the upregulation of HIF1A and BNIP3-mediated mitophagy. Autophagy 17:2975-2990.

Lin Q, Li S, Jiang N *et al*., 2019. PINK1-parkin pathway of mitophagy protects against contrast-induced acute kidney injury via decreasing mitochondrial ROS and NLRP3 inflammasome activation. Redox Biol 26: 101254.

Ma SQ, Guo Z, Liu FY *et al*., 2021. 6-Gingerol protects against cardiac remodeling by inhibiting the p38 mitogen-activated protein kinase pathway. Acta Pharmacol Sin 42:1575-1586.

Oh CJ, Kim MJ, Lee JM *et al*., 2023. Inhibition of pyruvate dehydrogenase kinase 4 ameliorates kidney ischemia-reperfusion injury by reducing succinate accumulation during ischemia and preserving mitochondrial function during reperfusion. Kidney Int 104:724-739.

Ostermann M, Lumlertgul N, Jeong R *et al*., 2025. Acute kidney injury. Lancet 405:241-256.

Pickkers P, Darmon M, Hoste E *et al*., 2021. Acute kidney injury in the critically ill: an updated review on pathophysiology and management. Intensive Care Med 47:835-850.

Pu JL, Huang ZT, Luo YH *et al*., 2021. Fisetin mitigates hepatic ischemia-reperfusion injury by regulating GSK3β/AMPK/NLRP3 inflammasome pathway. Hepatobiliary Pancreat Dis Int 20:352-360.

Rewa O, Bagshaw SM, 2014. Acute kidney injury-epidemiology, outcomes and economics. Nat Rev Nephrol 10: 193-207.

Rodrigues MJ, 2024. Bioprospecting of natural products from medicinal plants. Plants (Basel) 13:3556

Ronco C, Bellomo R, Kellum JA, 2019. Acute kidney injury. Lancet 394:1949-1964.

Segev G, Cortellini S, Foster JD *et al*., 2024. International Renal Interest Society best practice consensus guidelines for the diagnosis and management of acute kidney injury in cats and dogs. Vet J 305:106068.

Tang TT, Lv LL, Pan MM *et al*., 2018. Hydroxychloroquine attenuates renal ischemia/reperfusion injury by inhibiting cathepsin mediated NLRP3 inflammasome activation. Cell Death Dis 9:351.

Tsunekawa N, Sato M, 2024. Efficacy of oral AB070597 for the management of chronic kidney disease in cats: a prospective, randomised, controlled parallel-group study. J Feline Med Surg 26:1098612x241275249.

Wang S, Chen Y, Han S *et al*., 2022. Selenium nanoparticles alleviate ischemia reperfusion injury-induced acute kidney injury by modulating GPx-1/NLRP3/Caspase-1 pathway. Theranostics 12:3882-3895.

Wu S, Zhu J, Wu G *et al*., 2022. 6-Gingerol alleviates ferroptosis and inflammation of diabetic cardiomyopathy via the Nrf2/HO-1 pathway. Oxid Med Cell Longev 2022:3027514.

Yu Q, Li J, Cui M *et al*., 2024. 6-Gingerol attenuates hepatic ischemia/reperfusion injury through regulating MKP5-mediated P38/JNK pathway. Sci Rep 14:7747.

Zhu Q, Hao H, Gao Y *et al*., 2025. Dapagliflozin ameliorates kidney injury following limb ischemia-reperfusion via the AMPK/SIRT1/NLRP3 pathway. Ren Fail 47:2495111.