Endogenous SO\(_2\) Controls Cell Apoptosis: The State-of-the-Art

Yingying Li\(^1,2\)\(^\dagger\), Yingjun Feng\(^2\)\(^\dagger\), Xiaoyun Ye\(^1\), Hanlin Peng\(^1\), Jiantong Du\(^3\), Xiaoli Yao\(^2\), Yaqian Huang\(^1\)\(^*\), Hongfang Jin\(^1\)\(^*\) and Junbao Du\(^1,4\)\(^*\)

\(^1\) Department of Pediatrics, Peking University First Hospital, Beijing, China, \(^2\) Department of Cardiovascular Medicine, Children’s Hospital Affiliated to Zhengzhou University/Children’s Hospital of Henan Province, Zhengzhou, China, \(^3\) Department of Ophthalmology, Peking University First Hospital, Beijing, China, \(^4\) Key Lab of Molecular Cardiology, Ministry of Education, Beijing, China

SO\(_2\), previously known as the product of industrial waste, has recently been proven to be a novel gasotransmitter in the cardiovascular system. It is endogenously produced from the metabolism pathway of sulfur-containing amino acids in mammalians. Endogenous SO\(_2\) acts as an important controller in the regulation of many biological processes including cardiovascular physiological and pathophysiological events. Recently, the studies on the regulatory effect of endogenous SO\(_2\) on cell apoptosis and its pathophysiological significance have attracted great attention. Endogenous SO\(_2\) can regulate the apoptosis of vascular smooth muscle cells, endothelial cells, cardiomyocytes, neuron, alveolar macrophages, polymorphonuclear neutrophils and retinal photoreceptor cells, which might be involved in the pathogenesis of hypertension, pulmonary hypertension, myocardial injury, brain injury, acute lung injury, and retinal disease. Therefore, in the present study, we described the current findings on how endogenous SO\(_2\) is generated and metabolized, and we summarized its regulatory effects on cell apoptosis, underlying mechanisms, and pathophysiological relevance.

Keywords: sulfur dioxide, metabolism, apoptosis, mechanism, pathophysiology

INTRODUCTION

Previously, SO\(_2\) was recognized as a water-soluble, colorless, transparent exhaust gas and air pollutant with a sharp odor. High concentrations of SO\(_2\) in the environment could cause various degrees of damage to humans, animals, plants, and even microorganisms. However, it was discovered that SO\(_2\) can be endogenously produced in the mammalian metabolism of sulfur-containing amino acids (SAAs) (Stipanuk, 2004; Kimura, 2011). Many studies have shown that endogenous SO\(_2\) has the unique characteristics of continuous production, rapid diffusion, low molecular weight, and extensive action (Du et al., 2008a; Huang et al., 2016). More interestingly, it was found to play an important role in regulating cardiovascular function and structure under physiologic and pathophysiologic conditions (Du et al., 2008a; Huang et al., 2016). Therefore, we proposed that SO\(_2\) is a new cardiovascular gaseous signaling molecule. However, the molecular mechanisms underlying its biological action require further studies. It has been found recently that endogenous SO\(_2\) regulates cell apoptosis, and therefore contributes to the pathogenesis of hypertension, pulmonary hypertension, myocardial injury, brain injury, retinal disease, and acute lung injury (Zhao et al., 2008, 2013, 2019; Wang et al., 2011; Ma et al., 2012; Jin et al., 2013; Han et al., 2014; Du et al., 2018; Liu et al., 2018; Niu et al., 2018; Yang et al., 2018; Zhou et al., 2020). In this article, we described the generation and metabolism of endogenous SO\(_2\), and summarized...
the latest findings of the regulation of endogenous SO\textsubscript{2} on cell apoptosis, its mechanisms, and pathophysiological significance.

**GENERATION AND METABOLISM OF ENDOGENOUS SO\textsubscript{2}**

SO\textsubscript{2} can be produced endogenously through the biotransformation of SAAs in various mammal systems (Figure 1), such as the cardiovascular, respiratory, nervous, digestive, urinary, and immune systems. In the body, SAAs, such as methionine and cysteine, are primarily metabolized to L-cysteine (L-Cys). L-Cys is a crucial precursor of endogenous SO\textsubscript{2} (Singer and Kearney, 1956; Stipanuk, 2004; Du et al., 2008a; Kimura, 2011; Huang et al., 2016). It can be oxidized to produce L-cysteinesulfinate, and this process involves a key enzyme cysteine dioxygenase. Subsequently, the metabolism of L-cysteinesulfinate involves two pathways. In one pathway, L-cysteinesulfinate is converted into \( \beta \)-sulfanylpuruvate via the catalysis of aspartate aminotransferase (AAT), which is followed by spontaneous decomposition of \( \beta \)-sulfanylpuruvate into SO\textsubscript{2} and pyruvic acid. In the other pathway, L-cysteinesulfinate is decarboxylated into hypotaurine and CO\textsubscript{2} via cysteinesulfinate decarboxylase. Hypotaurine is subsequently oxidized to taurine. There is a competition between these two pathways for the reaction tendency of L-cysteinesulfinate, which is ultimately determined by the activities of AAT and cysteinesulfinate decarboxylase (Singer and Kearney, 1956; Griffith, 1983). Studies have indicated that \( 3' \)-phosphoadenosine-5'-phosphosulfate in activated neutrophils functions as an endogenous sulfate donor (Stipanuk, 2004).

Besides, hydrogen sulfide (H\textsubscript{2}S), another gaseous signaling molecule, metabolized from SAAs catalyzed by cystathionine \( \gamma \)-lyase or cystathionine \( \beta \)-synthase (Stipanuk, 2004; Kimura, 2011), can be transformed into SO\textsubscript{2} or sulfate through the following two pathways: (1) H\textsubscript{2}S is oxidized directly to sulfate or SO\textsubscript{2} through reduced nicotinamide adenine dinucleotide phosphate (NADPH) oxidase (Mitsuhashi et al., 2005). (2) H\textsubscript{2}S is firstly oxidized by sulfide oxidase into thiosulfate, and then the enzymes such as thiosulfate sulfurtransferase or glutathione-dependent thiosulfate reductase catalyze the transformation of thiosulfate to SO\textsubscript{2} (Shapiro, 1977).

In mammals, SO\textsubscript{2} can be hydrated to sulfite, and then converted into sulfate by sulfite oxidase, finally excreted in the urine in the kidney (Stipanuk, 2004; Huang et al., 2016). The half-life of SO\textsubscript{2} was supposed to be about 5–10 min, represented by the fact that serum SO\textsubscript{2} level decreased by 50% about 5–10 min after a bolus intravenous injection of SO\textsubscript{2} donor (Du et al., 2008b). It is known that AAT has two isozymes: cytosol AAT1 and mitochondrial AAT2. Huang et al. found that the overexpression of AAT1 or AAT2 increased the knockdown of AAT1 or AAT2 inhibited the endogenous SO\textsubscript{2} production, suggesting that AAT1/2 is the main SO\textsubscript{2}-generating enzyme (Liu et al., 2014).

SO\textsubscript{2} was detected in the headspace gas collected from a closed vial culturing porcine coronary arteries in 2003 by Balazy et al. (2003). Du et al. firstly found the SO\textsubscript{2}/AAT pathway in the cardiovascular tissues of rats in 2008 (Du et al., 2008a). The SO\textsubscript{2} level in rat plasma was 15.54 ± 1.68 \( \mu \text{mol/L} \) while the SO\textsubscript{2} concentration in the aorta, pulmonary artery, mesenteric artery, tail artery, and renal artery were 3.27 ± 0.21, 2.67 ± 0.17, 2.50 ± 0.20, and 2.23 ± 0.19 \( \mu \text{mol/g} \) protein, respectively. To date, SO\textsubscript{2}/AAT pathway has been found in almost all organs in mammals including vessels, heart, stomach, lung, liver, kidney, brain, retina, pancreas, and spleen, etc. (Luo et al., 2011; Du et al., 2018). It was found that the SO\textsubscript{2} level was the highest in the stomach, followed by the ventricle, and the activity of AAT and the expression of AAT1 mRNA are the highest in the left ventricle, whereas the expression of the AAT1 protein is the highest in the right ventricle. Furthermore, the expression of AAT2 mRNA is the highest in the liver, whereas the expression of the AAT2 protein is the highest in the renal medullary membrane (Luo et al., 2011).

**MECHANISMS BY WHICH SO\textsubscript{2} REGULATES CELL APOPTOSIS AND THE SIGNIFICANCE**

Apoptosis is the programmed cell death that occurs in multicellular organisms. Increasing evidence implies that endogenous SO\textsubscript{2} participates in regulating cell apoptosis (Zhao et al., 2008, 2013, 2019; Wang et al., 2011; Ma et al., 2012; Jin et al., 2013; Han et al., 2014; Du et al., 2018; Liu et al., 2018; Niu et al., 2018; Yang et al., 2018; Zhou et al., 2020, Figure 2). Three main pathways associated with the regulation of SO\textsubscript{2} on cell apoptosis have been revealed to date: (1) Extrinsic pathway (Wajant, 2002; Zhao et al., 2008): The combination of Fas ligand (FasL)/Fas causes the binding of the Fas-associated death domain proteins which activate caspase-8 precursor (caspase-10 in humans), initiate the cascade activation of caspase-3, caspase-6, and caspase-7 and then induce cell apoptosis. (2) Intrinsic pathway (Jin et al., 2013): The leakage of cytochrome c (CytC) from impaired mitochondria to cytosol is the central event of the mitochondria-related intrinsic apoptosis pathway. Its preceding events include the deformation and swelling of mitochondria, the imbalance of apoptotic regulator B-cell lymphoma-2 (bcl-2)/bcl-2-associated X protein (bax), the destroy of mitochondrial membrane potential (MMP), and the opening of mitochondrial permeability transition pore (MPTP). The cytosol CytC activates caspase-9 and caspase-3, finally induces cell apoptosis. (3) Endoplasmic reticulum stress (ERS)-related apoptosis (Wang et al., 2011): The overactivated ERS induces cell apoptosis via overexpressing C/EBP homologous protein (CHOP) and activating caspase-12.

**Endogenous SO\textsubscript{2} Promotes Vascular Smooth Muscle Cell Apoptosis**

As the main component cell of the vessel, the vascular smooth muscle cell (VSMC) apoptosis markedly affects vascular remodeling. Zhao et al. (2008) found that compared with the WKY rat, the plasma SO\textsubscript{2} level and the proportion of apoptotic VSMC in media were downregulated in the spontaneously hypertensive rat (SHR). Correspondingly, the blood pressure and...
the ratio of aorta media to lumen radius were increased by 53.6 and 28.1% in the SHR rat, respectively. Furthermore, SO\textsubscript{2} the ratio of aorta media to lumen radius were increased by Li et al. Endogenous SO\textsubscript{2} group. These data suggested that SO\textsubscript{2} associated with the different experimental conditions such as in vitro without fetal bovine serum stimulation (Liu et al., 2014). The discrepant effects of SO\textsubscript{2} in vitro remodeling by promoting VSMC apoptosis, which involved the regulation of bcl-2, Fas, and caspase-3. This might inhibit the hypoxia-induced HPAEC apoptosis by increasing bcl-2 expression, and inactivating caspase-3. by cyclic mechanical stretch and hydrostatic pressure and indirectly affected by endothelial cell (EC)-transduced shear stress under in vivo circumstances (Qiu et al., 2013; Chen et al., 2021). The abovementioned biomechanical stresses are reported to control the VSMC apoptosis, proliferation, and other behaviors and only are partly mimicked in vivo.

### Endogenous SO\textsubscript{2} Inhibits Vascular Endothelial Cell Apoptosis
Vascular ECs constitute a huge divider separating the lumen and wall of the vessel, and therefore play a crucial role in maintaining the vascular function and microenvironment homeostasis. Especially, vascular EC apoptosis is a key process involved in the vascular physiological and pathophysiological regulation such as organ development, angiogenesis, vascular injury, and remodeling (Duan et al., 2021; Tisch and Ruiz de Almodóvar, 2021). During the development of hypoxic pulmonary artery structural remodeling, hypoxia-induced vascular EC apoptosis is a critical pathological change. Liu et al. explored the effect of SO\textsubscript{2} on the human pulmonary artery endothelial cell (HPAEC) apoptosis based on cobalt chloride (CoCl\textsubscript{2})-stimulated cell model (Liu et al., 2018). The endogenous SO\textsubscript{2} production and AAT1 expression were down-regulated but the percentage of terminal deoxynucleotidyl transferase-mediated nick end labeling (TUNEL)-positive nuclei was increased in HPAECs of the vehicle + CoCl\textsubscript{2} group as compared with those of the vehicle group. However, there was no difference in the endogenous SO\textsubscript{2}/AAT1 pathway and apoptosis of HPAECs between the AAT1 overexpression and AAT1 overexpression + CoCl\textsubscript{2} groups, suggesting that the sufficient endogenous SO\textsubscript{2} was a powerful antagonist to chemical hypoxia-induced HPAEC apoptosis. Furthermore, AAT1 overexpression prevented the downregulation of antiapoptotic factor bcl-2, and the activation of caspase-3 in the CoCl\textsubscript{2}-treated HPAECs. These data suggested that the endogenous SO\textsubscript{2} might inhibit the hypoxia-induced HPAEC apoptosis by increasing bcl-2 expression, and inactivating caspase-3.

### Endogenous SO\textsubscript{2} Regulates Cardiomyocyte Apoptosis
Cardiomyocyte apoptosis is a keystone of cardiac pathological changes and is closely correlated with the development of heart failure, myocardial ischemia-reperfusion (I/R) injury and cardiomyopathy, etc. Endogenous SO\textsubscript{2} was found to protect against the myocardial I/R, isopropylationerol (ISO)-induced cardiac injury, sepsis-induced cardiac dysfunction, myocardial infarction, and diabetic cardiomyopathy, suggesting that cardiomyocyte might be an important target cell of endogenous SO\textsubscript{2} (Liang et al., 2011; Wang et al., 2011, 2018; Chen et al., 2012; Jin et al., 2013; Zhao et al., 2013; Liu et al., 2017; Yang et al., 2018; Zhou et al., 2020). SO\textsubscript{2} preconditioning significantly reduced the size of the myocardial infarction and decreased the percentage of TUNEL-positive cells in the rats of myocardial I/R (Wang et al., 2011). The mechanisms by which SO\textsubscript{2} preconditioning inhibited the cardiomyocyte apoptosis in the myocardial I/R

![FIGURE 1](https://www.frontiersin.org) Production and metabolism of endogenous SO\textsubscript{2} in mammals. L-Cys is metabolized through important enzymes including CDO, AAT, and CSD to generate endogenous SO\textsubscript{2}, pyruvic acid, and hypotaurine. Moreover, H\textsubscript{2}S, a product of the reaction catalyzed by CSE and CBS with L-Cys as substrate, can be metabolized to sulfite or SO\textsubscript{2} via the catalyzation of sulfide oxidase, TST, and NADPH oxidase. Thirdly, PAPS in activated neutrophils is also an origin of sulfite. In mammals, SO\textsubscript{2} can be hydrated to sulfite, then converted into sulfate by sulfite oxidase, and finally excreted in the urine in the kidney. AAT: aspartate aminotransferase; CBS, cystathionine \(\beta\)-synthase; CDO, cysteine dioxygenase; CSE, cystathionine \(\gamma\)-lyase; L-Cys, L-cysteine; NADPH, nicotinamide dinucleotide phosphate; PAPS, \(3'\)-phosphoadenosine-\(5'\)-phosphosulfate; TST, thiosulfate sulfurtransferase.
FIGURE 2 | The effect of endogenous SO$_2$ on cell apoptosis. According to the clockwise direction, the SO$_2$ target cells are shown beginning from vascular smooth muscle cells. In the cardiovascular system, endogenous SO$_2$ promotes vascular smooth muscle cell apoptosis, inhibits vascular endothelial cell apoptosis, and inhibits/promotes cardiomyocyte apoptosis. In the nervous system, endogenous SO$_2$ inhibits/promotes neuron apoptosis, and reduces the apoptosis of retinal photoreceptor cells. Regarding immune cells, SO$_2$ promotes polymorphonuclear neutrophil apoptosis but inhibits macrophage apoptosis.

might involve the following (Wang et al., 2011; Zhao et al., 2013): (1) SO$_2$ preconditioning induced a moderate ERS, and then inhibited vigorous ERS-initiated cardiomyocyte apoptosis in the development of myocardial I/R injury. The above process was supported by the fact that SO$_2$ induced early ERS markers glucose-regulated protein 78 (Grp78) expression and eukaryotic initiation factor 2a (eIF2a) phosphorylation, while myocardial I/R induced early and late ERS markers including Grp78 expression, eIF2a phosphorylation, CHOP expression and caspase-12 activation, which could be alleviated by SO$_2$ preconditioning. (2) SO$_2$ preconditioning inhibited the activation of caspase-9 and caspase-3 in the rats of the myocardial I/R group. Considering that caspase-9 activation subsequently follows the leakage of cytosol Cytc from injury mitochondria, the antiapoptotic effect of SO$_2$ preconditioning might involve mitochondrial protection. (3) The inhibitor of PI3K/Akt the pathway blocked the SO$_2$ preconditioning-inhibited caspase-3 activation, suggesting that the activation of the PI3K/Akt pathway might mediate the protective role of SO$_2$ preconditioning.

The effect of endogenous SO$_2$ on the ISO-induced cardiomyocyte apoptosis was examined targeting the mitochondrion (Liang et al., 2011; Jin et al., 2013). The SO$_2$ level in the myocardial tissue was decreased but the proportion of apoptotic cells was increased in the rats of the ISO group compared with those of the control group. SO$_2$ treatment restored the myocardial SO$_2$ level and inhibited cardiomyocyte apoptosis. Then, the study further focused on the mitochondria. Firstly, the stereological analysis of the mitochondria showed that SO$_2$ alleviated the cardiomyocytic mitochondria swelling and deformation, represented by the fact that SO$_2$ treatment decreased the mitochondrial mean surface area, mean volume and volume density, and increased the mitochondrial numerical density and surface-to-volume ratio in the myocardial tissue of ISO-treated rats. Secondly, SO$_2$ upregulated bcl-2 expression and downregulated bax expression, improved the impaired MMP, and reclosed the MPTP in rat myocardial tissues. Subsequently, SO$_2$ blocked the leakage of Cytc from the mitochondria and inhibited the activation of cytosol caspase-9 and caspase-3. These data suggested that endogenous SO$_2$ prevented ISO-induced cardiomyocytic apoptosis via maintaining the mitochondrial function and structure. Moreover, SO$_2$ decreased the protein and mRNA levels of ERS markers Grp78, CHOP, and caspase-12 in the myocardial tissues of ISO-treated rats, suggesting that the over-activated ERS might also be involved in the inhibitory effect of SO$_2$ on the ISO-induced cardiomyocytic apoptosis (Chen et al., 2012).

Similar to the ISO-induced myocardial injury, SO$_2$ treatment inhibited cardiomyocytic apoptosis in the myocardial tissue of rats with cecal ligation and puncture (CLP), accompanied by an increase in the bcl-2 expression, and a decrease in the bax/bcl-2 ratio and caspase-3 activation (Yang et al., 2018). Moreover, SO$_2$ decreased the level of inflammatory cytokines, inhibited TLR4 and NLRP3 expression, and reduced the inflammatory
Endogenous SO\textsubscript{2} Regulates Neuronal Apoptosis

The relationship between endogenous SO\textsubscript{2} and neuronal apoptosis in the neurological disease was investigated on the kainic acid (KA)-induced epilepsy, febrile seizure (FS), and transient global ischemia/reperfusion models (Han et al., 2014; Niu et al., 2018; Zare Mehrjerdi et al., 2018). The different effects of SO\textsubscript{2} on neuronal apoptosis depended on the neurological disorders and SO\textsubscript{2} donor concentrations.

In the KA-induced epileptic rats, plasma SO\textsubscript{2} level and AAT activity were upregulated compared with those in control rats (Niu et al., 2018). However, an AAT inhibitor HDX treatment retarded the occurrence of hippocampal neuronal apoptosis in the KA-treated rats, represented by the fact that a significant hippocampal neuronal apoptosis was detected in the rats 72 h after treatment with KA, while it was not observed in the rats treated with KA + HDX until 7 days after the intervention. The results suggested that the upregulated endogenous SO\textsubscript{2} might promote hippocampal neuronal apoptosis in the development of epilepsy-related brain injury. Furthermore, the KA-upregulated ERS markers Grp78 expression and phosphorylated PERK in the hippocampus could be inhibited by HDX, indicating that ERS might participate in the promotive effect of SO\textsubscript{2} on hippocampal neuronal apoptosis.

The endogenous SO\textsubscript{2}/AAT pathway including SO\textsubscript{2} content and AAT1/2 expressions was upregulated in the hippocampus in rats of recurrent FS, accompanied by an increase in the neuronal apoptosis and mossy fiber sprouting (MFS) in the hippocampus (Han et al., 2014). Furthermore, HDX treatment aggravated the increased neuronal apoptosis and MFS in recurrent FS rats, while the supplement of a low concentration of SO\textsubscript{2} (1–10 µmol/kg) alleviated the above pathological change, suggesting that the upregulated endogenous SO\textsubscript{2} might be a defensive mechanism for antagonizing the neuronal apoptosis in the development of recurrent FS. However, the supplement of a high concentration of SO\textsubscript{2} donor (100 µmol/kg) worsened the neuronal apoptosis and MFS in recurrent FS rats, which reminded us to pay attention to the protective dose range of SO\textsubscript{2}.

A global brain I/R injury increased CA1 neuronal apoptosis in the rats undergoing the occlusion of both common carotid arteries, accompanied by an increase in malondialdehyde level, and a decrease in glutathione level and activity of superoxide dismutase in the CA1 region (Zare Mehrjerdi et al., 2018). Furthermore, SO\textsubscript{2} donor treatment improved the learning and memory deficits in the rats with cerebral I/R injury, reduced CA1 neuronal apoptosis, decreased malondialdehyde level, and elevated GSH level and activity of superoxide dismutase in the CA1 region. These findings indicated that SO\textsubscript{2} donor prevented brain ischemic injury by decreasing neuronal apoptosis and the underlying mechanisms might involve the anti-oxidative effect of SO\textsubscript{2}. However, the change of endogenous SO\textsubscript{2}/AAT pathway in the brain ischemic rats was not detected, which merits further studies for exploring the role of endogenous SO\textsubscript{2} in the development of brain ischemic injury.

Endogenous SO\textsubscript{2} Inhibits Retinal Photoreceptor Cell Apoptosis

Oxidative stress can induce apoptosis of retinal photoreceptor cells, which is the main pathological change of blinding retinal diseases (Wenzel et al., 2005). Du et al. found that apoptosis was significantly induced in the 661w mouse photoreceptor cells exposed to H\textsubscript{2}O\textsubscript{2} compared with those without H\textsubscript{2}O\textsubscript{2} exposure, demonstrated by an increase in the percentage of TUNEL positive cells and the ratio of cleaved-caspase3/caspase3 (Du et al., 2018). Simultaneously, endogenous SO\textsubscript{2} production and AAT1 expression were decreased in the H\textsubscript{2}O\textsubscript{2}-treated 661w cells compared with control cells. HDX treatment mimicked the effects of H\textsubscript{2}O\textsubscript{2} on 661w cells including an increase in the cell apoptosis and the downregulation of endogenous SO\textsubscript{2}/AAT pathway, indicating that the apoptosis of 661w cells caused by oxidative stress might be mediated by the downregulation of the endogenous SO\textsubscript{2} pathway. However, the role of endogenous SO\textsubscript{2} in the development of retinal photoreceptor cell apoptosis and its mechanisms require further studies.

Endogenous SO\textsubscript{2} Promotes Polymorphonuclear Neutrophil Apoptosis

Lipopolysaccharide (LPS) was intratracheally instilled to induce acute lung injury (ALI) in adult male rats (Huang et al., 2009). The decrease in the apoptosis of polymorphonuclear neutrophils (PMNs) contribute to an increment of the number of PMN and a prolonged inflammatory reaction in the injured lung (Lee and Downey, 2001). LPS treatment reduced the SO\textsubscript{2} level in the lung tissue and peripheral blood, while the treatment of SO\textsubscript{2} donor alleviated the lung histopathological changes in the rats accompanied by a reduction of PMN number in the bronchoalveolar lavage fluid (Huang et al., 2009; Ma et al., 2012). Furthermore, SO\textsubscript{2} promoted the apoptosis of PMN in the bronchoalveolar lavage fluid and peripheral blood in the ALI...
rats. Mechanistically, SO$_2$ treatment upregulated the expression of bax, downregulated the expression of bcl-2, and elevated the level of activated caspase-3 in the peripheral PMN in the ALI rats. Therefore, it is believed that SO$_2$ may antagonize ALI by promoting PMN apoptosis, in which the bax/bcl-2/caspase-3 pathway is involved.

**Endogenous SO$_2$ Inhibits Alveolar Macrophage Apoptosis**

Many studies focused on the role of alveolar macrophage (AM) in the pathogenesis of ALI (Cheng et al., 2021). It was found that AM apoptosis was significantly increased in the ALI rat model induced by limb I/R injury, resulting in the delayed clearance of inflammatory PMN and the prolongation of ALI (Zhao et al., 2016). Zhao et al. found that SO$_2$ donor prevented limb I/R-induced ALI in rats (Zhao et al., 2016). Furthermore, the primary AM was cultured with the plasma extracted from limb I/R to make the limb I/R-induced AM injury model (Zhao et al., 2019). Compared with the ALI group, the AM apoptosis was reduced in the AMs of the ALI + SO$_2$ group, with an increase in bcl-2 expression, an improvement of the impaired MPP, the reclosing of MPTP and the decrease in caspase-3 expression. These data suggested that the SO$_2$-inhibited AM apoptosis might be involved in the protective effect of SO$_2$ on ALI and mitochondrion was the possible target of SO$_2$.

**CONCLUSION AND PERSPECTIVE**

As a novel gaseous signaling molecule, endogenous SO$_2$ was reported to play a variety of biological effects by regulating cell apoptosis. In summary, endogenous SO$_2$ promoted VSMC apoptosis and inhibited vascular EC apoptosis to alleviate the vascular remodeling in hypertension and pulmonary hypertension. Endogenous SO$_2$ suppressed the apoptosis of cardiomyocytes, contributing to the cardioprotective effect of SO$_2$. In the nervous system, SO$_2$ increased KA-induced hippocampal neuronal apoptosis to aggravate the epileptic brain damage, while SO$_2$ reduced neuronal apoptosis to protect against the neuronal damage in the recurrent FS and global brain I/R injury. Moreover, the downregulation of endogenous SO$_2$ was accompanied by the increase in the 661w retinal photoreceptor cell apoptosis, suggesting that endogenous SO$_2$ might participate in the pathogenesis of blinding retinal disease. In the respiratory system, SO$_2$ facilitated PMN apoptosis and inhibited AM apoptosis to protect against ALI. The abovementioned apoptosis-related studies proved that the abnormal endogenous SO$_2$ was involved in the pathogenesis of many diseases such as cardiovascular disease, neurological disease, and respiratory disease, and then keeping endogenous SO$_2$ in the normal range might be a new and meaningful therapeutic strategy of related diseases in the future.

However, there remain some issues to be taken into account in future studies: (1) The endogenous SO$_2$ generation and its producing enzyme should be firstly detected in the animal and cell models to identify the role of endogenous SO$_2$ in the pathogenesis of diseases. (2) The concentration and type of SO$_2$ donors might affect the effect of SO$_2$, which should be paid attention to *in vitro* and *in vivo*. (3) Since mitochondria and their related apoptosis pathways were reported to be involved in almost all the anti- or pro-apoptotic effects of endogenous SO$_2$, the detailed target molecules, and mechanisms by which SO$_2$ affects mitochondria merit expectation in the future. (4) A recent study showed useful findings that plasma SO$_2$ as a practical biomarker could predict the occurrence of acute kidney injury in the patients staying in the surgical intensive care unit (Jiang et al., 2021). Therefore, as a biomarker, the level of SO$_2$ in clinical samples might have translational potential in the diagnosis, treatment decision, and prognosis prediction of the diseases. (5) In the future, the SO$_2$-released prodrug and AAT inhibitor in clinical application for apoptosis-related diseases are expected to further studies (Day et al., 2016; Wang and Wang, 2018; Huang et al., 2021).

**AUTHOR CONTRIBUTIONS**

All authors listed have made a substantial, direct and intellectual contribution to the work, and approved it for publication.

**FUNDING**

This research was funded by National Natural Science Foundation of China (81770422 to YH, 81770278 to JBD, and 81900872 to JTD), Beijing Natural Science Foundation (7191012 to HJ).

**REFERENCES**

Balazy, M., Abu-Yousef, I. A., Harpp, D. N., and Park, J. (2003). Identification of carbonyl sulfide and sulfur dioxide in porcine coronary artery by gas chromatography/mass spectrometry, possible relevance to EDHF. *Biochem. Biophys. Res. Commun.* 331, 728–734. doi: 10.1016/j.bbrc.2003.10.055

Chen, S., Du, J., Liang, Y., Ochs, T., Liu, D., Zhu, L., et al. (2012). Sulfur dioxide inhibits excessive activated endoplasmic reticulum stress in rats with myocardial injury. *Heart Vessels* 27, 505–516. doi: 10.1007/s00380-011-0192-7

Chen, Z., Zhang, H., Bai, Y., Cui, C., Li, S., Wang, W., et al. (2021). Single cell transcriptomic analysis identifies novel vascular smooth muscle subsets under high hydrostatic pressure. *Sci. China Life Sci.* doi: 10.1007/s11427-020-1852-x [Epub ahead of print].

Cheng, P., Li, S., and Chen, H. (2021). Macrophages in lung injury, repair, and fibrosis. *Cells* 10:436. doi: 10.3390/cells10020436

Day, J. J., Yang, Z., Chen, W., Pacheco, A., and Xian, M. (2016). Benzothiazole sulfinate: a water-soluble and slow-releasing sulfur dioxide donor. *ACS Chem. Biol.* 11, 1647–1651. doi: 10.1021/acschembio.6b00106

Du, J., Huang, Y. Q., Li, K., Yu, X. Q., Jin, H. F., and Yang, L. (2018). Retina-derived sulfuric acid might be a novel anti-apoptotic factor. *Biochem. Biophys. Res. Commun.* 496, 955–960. doi: 10.1016/j.bbrc.2018.01.103

Du, S. X., Jin, H. F., Bu, D. F., Zhao, X., Geng, B., Tang, C. S., et al. (2008a). Endogenously generated sulfur dioxide and its vasorelaxant effect in rats. *Acta Pharmacol. Sin.* 29, 923–930. doi: 10.1111/j.1745-7254.2008.08845.x

Du, S. X., Jin, H. F., Liang, Y. F., Zhao, X., Wei, H. L., Wang, L., et al. (2008b). Influence of sulfur dioxide and its derivatives on rats’ blood pressure. *J. Appl. Clin. Pediatr.* 23, 22–24. doi: 10.3969/j.issn.1000-515X.2008.01.007
Duan, H., Zhang, Q., Liu, J., Li, R., Wang, D., Peng, W., et al. (2021). Suppression of apoptosis in vascular endothelial cell, the promising way for natural medicines to treat atherosclerosis. Pharmacol. Res. 186:105599. doi: 10.1016/j.phrs.2021.105599

Griffith, O. W. (1983). Cysteinesulfinate metabolism: altered partitioning between transamination and decarboxylation following administration of beta-methylenecaptopitate. J. Biol. Chem. 258, 1591–1598.

Han, Y., Yi, W., Qin, J., Zhao, Y., Zhang, J., and Chang, X. (2014). Dose-dependent effect of sulfur dioxide on brain damage induced by recurrent febrile seizures in rats. Neurosci. lett. 563, 149–154. doi: 10.1016/j.neulet.2013.12.042

Huang, X. L., Zhou, J. L., Zhou, X. H., Xian, X. H., and Ding, C. H. (2009). Ameliorative effects of exogenous sulfur dioxide on lipopolysaccharide-induced acute lung injury in rats. Acta Physiol. Sin. 61, 499–503. doi: 10.3321/j.issn:0371-0874.2009.05.014

Huang, Y. Q., Tan, C. X., Du, J. B., and Jin, H. F. (2016). Endogenous sulfur dioxide: a new gasotransmitter in the cardiovascular system. Oxid. Med. Cell. Longev. 2016:896195. doi: 10.1155/2016/896195

Huang, Y. Q., Zhang, H., Lv, B. Y., Tang, C. S., Du, J. B., and Jin, H. F. (2021). Endogenous sulfur dioxide is a new gasotransmitter with promising therapeutic potential in cardiovascular system. Sci. Bull. 66, 1604–1607. doi: 10.1007/s10000-021-0138-7

Jiang, Y., Wang, J., Zheng, X., and Du, J. (2021). Plasma endogenous sulfur dioxide: a novel biomarker to predict acute kidney injury in critically ill patients. Int. J. Gen. Med. 14, 2127–2136. doi: 10.2147/IJGM.S312058

Jin, H., Liu, A. D., Holmberg, L., Zhao, M., Chen, S., Yang, J., et al. (2013). The role of sulfur dioxide in the regulation of mitochondrion-related cardiomycyte apoptosis in rats with isopropylylaminedol-induced myocardial injury. Int. J. Mol. Sci. 14, 10465–10482. doi: 10.3390/ijms14051046

Kimura, H. (2011). Hydrogen sulfide: its production, release and functions. IUBMB Life 63, 831–836. doi: 10.1002/iub.563

Li, Y., Yi, W., Qin, J., Zhao, Y., Zhang, J., and Chang, X. (2014). Biogenic release of hydrogen sulfide induces delayed cardiomyocyte apoptosis in rats. Biochem. Biophys. Res. Commun. 441, 113–121. doi: 10.1016/j.bbrc.2014.04.023

Liu, M., Liu, S., Tan, W., Long, J., Li, Z., et al. (2017). Gaseous signalling molecules: the sulfur dioxide pathway and its role on cell apoptosis and injury in diabetes rats. Mol. Med. Rep. 26, 876–882. doi: 10.3892/mmr.2017.7714

Liang, Y., Liu, D., Ochs, T., Tang, C., Chen, S., Zhang, S., et al. (2011). Endogenous sulfur dioxide protects against isoproterenol-induced myocardial injury and increases myocardial antioxidant capacity in rats. Lab. Invest. 91, 12–23. doi: 10.1038/labinvest.2010.156

Liu, D., Huang, Y., Bu, D., Liu, A. D., Holmberg, L., Jia, Y., et al. (2014). Sulfur dioxide inhibits vascular smooth muscle cell proliferation via suppressing the ERK/MAP kinase pathway mediated by CAMPA/PKA signaling. Cell Death Dis. 5,e1251. doi: 10.1038/cddis.2014.229

Liu, M., Liu, S., Tan, W., Tang, F., Long, J., Li, Z., et al. (2017). Gaseous signalling molecule SO2 via Hippo-MST pathway to improve myocardial fibrosis of diabetic rats. Mol. Med. Rep. 16, 8935–8963. doi: 10.3892/mmr.2017.10200

Liu, X., Su, Z., Jin, X., Qin, G., and Nan, S. (2019). Sulfur dioxide induces apoptosis via reactive oxygen species generation in rat cardiomyocytes. Environ. Sci. Pollut. Res. 26, 8756–8767. doi: 10.1007/s11356-019-04319-7

Zhao, Y. R., Liu, Y., Wang, D., Lv, W. R., and Zhou, J. L. (2019). Effects of sulfur dioxide on alveolar macrophage apoptosis in acute lung injury induced by limb ischemia/reperfusion in rats. Acta Pharmacol. Sin. 40, 501–506. doi: 10.1038/aps.2018.204

Zhao, X., Jin, H. F., Tang, C. S., and Du, J. B. (2008). Effects of sulfur dioxide on hippocampal cell death and improves learning and memory deficits in a rat model of transient global ischemia/reperfusion. Iran. J. Basic Med. Sci. 21, 998–1003. doi: 10.22038/ijbms.2018.29404.7106

Zhao, M. M., Yang, J. Y., Wang, X. B., Tang, C. S., Du, J. B., and Jin, H. F. (2013). The PI3K/Akt pathway mediates the protection of SO2 preconditioning against myocardial ischemia/reperfusion injury in rats. Acta Pharmacol. Sin. 34, 501–506. doi: 10.1038/aps.2012.204

Zhao, X., Jin, H. F., Tang, C. S., and Du, J. B. (2008). Effects of sulfur dioxide, on the proliferation and apoptosis of aorta smooth muscle cells in hypertension: experiments with rats. Zhonghua Yi Xue Za Zhi 88, 1279–1283. doi: 10.3321/j.issn:0376-2491.2008.18.011

Zhao, Y. R., Liu, Y., Wang, D., Lv, W. R., and Zhou, J. L. (2019). Effects of sulfur dioxide on alveolar macrophage apoptosis in acute lung injury induced by limb ischemia/reperfusion in rats. J. Peking Univ. 51, 239–244. doi: 10.10723/jissn:1671-161X.2019.02.002

Zhao, Y. R., Du, D., Liu, Y., Shan, L., and Zhou, J. L. (2016). The PI3K/Akt/p38MAPK, and JAK2/STAT3 signaling pathways mediate the protection of SO2 against acute lung injury induced by limb ischemia/reperfusion in rats. J. Physiol. Sci. 66, 229–239. doi: 10.1007/s12576-015-0414-z

Conflict of Interest: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Publisher's Note: All claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article, or claim that may be made by its manufacturer, is not guaranteed or endorsed by the publisher.

Copyright © 2021 Li, Feng, Ye, Peng, Du, Yao, Huang, Jin and Du. This is an open-access article distributed under the terms of the Creative Commons Attribution License (CC BY). The use, distribution or reproduction in other forums is permitted, provided the original author(s) and the copyright owner(s) are credited and that the original publication in this journal is cited, in accordance with accepted academic practice. No use, distribution or reproduction is permitted which does not comply with these terms.