ScienceDirect

Biochemical and Biophysical Research Communications

Volume 495, Issue 2, 8 January 2018, Pages 1620-1627

Hydrogen protects against hyperoxia-induced apoptosis in type II alveolar epithelial cells via activation of PI3K/Akt/Foxo3a signaling pathway

Dan Wu [a b c], Mulin Liang [a b c], Hongxing Dang [a b c], Fang Fang [a b c], Feng Xu [a b c], Chengjun Liu [a b c]

Show more

https://doi.org/10.1016/j.bbrc.2017.11.193

Get rights and content

Abstract

Oxidative stress is regarded as a key regulator in the pathogenesis of prolonged hyperoxia-induced lung injury, which causes injury to alveolar epithelial cells and eventually leads to development of bronchopulmonary dysplasia (BPD). Many studies have shown that hydrogen has a protective effect in a variety of cells. However, the mechanisms by which hydrogen rescues cells from damage due to oxidative stress in BPD remains to be fully elucidated. This study sought to evaluate the effects of hydrogen on hyperoxia-induced lung injury and to investigate the underlying mechanism. Primary type II alveolar epithelial cells (AECIIs) were divided into four groups: control (21% oxygen), hyperoxia (95% oxygen), hyperoxia+hydrogen, and hyperoxia+hydrogen+LY294002 (a PI3K/Akt inhibitor). Proliferation and apoptosis of AECIIs were assessed using MTS assay and flow cytometry (FCM), respectively. Gene and protein expression were detected by quantitative polymerase chain reaction (q-PCR) and western blot analysis. Stimulation with hyperoxia decreased the expression of P-Akt, P- FoxO3a, cyclinD1 and Bcl-2. Hyperoxic conditions increased levels of Bim, Bax, and Foxo3a, which induced proliferation restriction and apoptosis of AECIIs. These effects of hyperoxia were reversed with hydrogen pretreatment. Furthermore, the protective effects of hydrogen were abrogated by PI3K/Akt inhibitor LY294002. The results indicate that hydrogen protects AECIIs from hyperoxia-induced apoptosis by inhibiting apoptosis factors and promoting the expression of anti-apoptosis factors. These effects were associated with activation of the PI3K/Akt/FoxO3a pathway.

Introduction

Oxygen therapy is commonly used in the treatment of respiratory diseases. However, prolonged exposure to high concentrations of oxygen can result in chronic pulmonary diseases and bronchopulmonary dysplasia (BPD) in neonates, especially preterm infants. Prolonged exposure to hyperoxic conditions can result in abnormal repair of lung tissue and pulmonary dysfunction after injury [1], [2]. Various clinical treatments, including stem cell transplantation, surfactant replacement therapy, corticosteroid administration, the long-term effect of treatment is not stable, more important is whether treatment will cause damage to other organ function is not clear [3], [4]. The proliferation and differentiation of type II alveolar epithelial cells (AECIIs) play a vital role in repair of alveolar structure and function in lung injury [5]. Therefore, a novel material is urgently needed to improve the viability and proliferation of AECII after exposure to hyperoxia.

Hydrogen has a selective antioxidant effect and neutralizes harmful free radicals [6], protects DNA and protein, preserves mitochondrial function, and prevents apoptosis. Molecular hydrogen protects against injury in several organs (including heart [7], lung [8], [9], bowel [10], eye [11], kidney [12], [13], brain [14], and pancreas [15]). Previous studies have shown that hydrogen can slow the development of BPD in newborn rats by reducing the release of inflammatory factors and reactive oxygen species (ROS) and the expression of inflammatory genes [16]. Preliminary experiments showed that hydrogen can significantly reduce oxidative damage to AECIIs and reduce apoptosis [17]. However, the underlying mechanisms of hydrogen's anti-apoptotic effects in the context of BPD remain elusive.

As one of the most important signal transduction pathways in the cell, PI3K-Akt signaling plays a pivotal role in promoting cell proliferation and inhibiting apoptosis through activation of downstream effector molecules [18], [19]. Transcription factor Forkhead box O3 (FoxO3a) is one of the most important downstream targets of PI3K/Akt signaling and a crucial regulator of cell apoptosis and cell-cycle arrest [20], A large number of reports have confirmed that inhibition of AKT promotes FOXO3a-dependent apoptosis in many cell types [21], [22], [23]. Previous research showed that hydrogen-rich saline treatment could attenuate neuronal apoptosis in early brain injury (EBI) and improve neurofunctional outcomes after subarachnoid hemorrhage (SAH), via activation of Akt/GSK3β signaling pathway [14]. However, whether hydrogen affects the expression and function the PI3K-Akt/Foxo3a pathway in hyperoxia-induced BPD remains largely unknown. The aim of the present study is to investigate whether hydrogen protects AECII from hyperoxia-induced injury by regulating the PI3K/AKT/Foxo3a signaling pathway.

Section snippets

Reagents

Pregnant Sprague-Dawley (SD) rats were purchased from Experimental Animal Center of the Daping Hospital of the Third Military Medical University (Chongqing, China). Trypsin, Dulbecco's modified Eagle's medium/Ham's Nutrient Mixture F12 (DMEM/F12) and fetal bovine serum (FBS) were purchased from Gibco (Invitrogen, USA). The annexin V FITC-labeled apoptosis detection kit and cell cycle assay kit were provided by Nanjing KeyGEN Biotech (Nanjing, China). MTS Cell Proliferation Colorimetric Assay…

Hydrogen protects AECIIs against hyperoxia-induced apoptosis via the PI3K/Akt pathway

To investigate the effects of hydrogen in protecting against hyperoxia–induced apoptosis, the viability and apoptosis rate of AECIIs was detected by MTS assay and Annexin V/PI (FCM), respectively. After exposure to hyperoxia, MTS results showed that cell viability was notably suppressed ($P<0.01$); hydrogen treatment markedly attenuated the cell growth inhibition caused by hyperoxia ($P<0.01$). However, cell viability was significantly decreased by co-treatment with hydrogen and LY294002…

Discussion

Oxidative damage represents the pathophysiological basis of most diseases. Continuous high-concentration oxygen therapy can cause excessive production of reactive oxygen species (ROS), which leads to pulmonary epithelial cell damage and diffuse alveolar injury. The alveolus is composed of type I alveolar epithelial cells (AECI) and AECII. AECIs are highly differentiated cells that have lost the capacity for regeneration. Therefore, structural and functional repair of alveolar epithelial injury…

Conflict of interest

The authors declare no conflict of interest.…

Acknowledgements

This study was supported by the National Natural Science Foundation of China (No. 30973218) and the project of foundation and frontier research program of Chongqing Science and Technology Commission (cstc2013jcyjA10031).…

References (35)

B.W. Buczynski *et al.*

The role of hyperoxia in the pathogenesis of experimental BPD

Semin. Perinatol. (2013)

H. Chen *et al.*

The effects of hydrogen-rich saline on the contractile and structural changes of intestine induced by ischemia-reperfusion in rats

J. Surg. Res. (2011)

H. Chi

miR-194 regulated AGK and inhibited cell proliferation of oral squamous cell carcinoma by reducing PI3K-Akt-FoxO3a signaling

Biomed. Pharmacother. (2015)

P. Yu *et al.*

Hydrogen-rich medium protects human skin fibroblasts from high glucose or mannitol induced oxidative damage

Biochem. Biophys. Res. Commun. (2011)

J. Chen *et al.*
Isolation of highly pure alveolar epithelial type I and type II cells from rat lungs
Lab. Invest. (2004)

T. Tanaka *et al.*
Cyclic mechanical stretch-induced oxidative stress occurs via a NOX-dependent mechanism in type II alveolar epithelial cells
Respir. Physiol. Neurobiol. (2017)

J. Cai *et al.*
Hydrogen therapy reduces apoptosis in neonatal hypoxia-ischemia rat model
Neurosci. Lett. (2008)

Y. Ozsurekci *et al.*
Oxidative stress related diseases in newborns
Oxid. Med. Cell Longev. (2016)

N.T. Liem *et al.*
Bone marrow mononuclear cells transplantation in treatment of established bronchopulmonary dysplasia: a case report
Am. J. Case Rep. (2017)

W. Onland *et al.*
Late (≥ 7 days) inhalation corticosteroids to reduce bronchopulmonary dysplasia in preterm infants
Cochrane Database Syst. Rev. (2017)

View more references

Cited by (50)

Effects of rapamycin and OSI-027 on α-SMA in lung tissue of SD rat pups with hyperoxic lung injury
2021, Biochemical and Biophysical Research Communications

Citation Excerpt :

…Fig. 2). This study adopted a mature animal model of hyperoxia lung injury established at our laboratory [11,12]. Pathological examination found that the whole process involved inflammatory reaction, and showed typical three-stage inflammation changes, namely, exudation, proliferation, and fibrosis.…

Show abstract

Isoleucine improved growth performance, and intestinal immunological and physical barrier function of hybrid catfish Pelteobagrus vachelli × Leiocassis longirostris

2021, Fish and Shellfish Immunology

> *Citation Excerpt :*
>
> ...Those data suggested that dietary Ile deficiency might influence cellular structural integrity associated with oxidative damage, cell apoptosis and dysfunction of TJs. The oxidative damage, cell apoptosis, and TJs could be regulated by NF-E2-related factor 2 (Nrf2) in human [33], protein kinase B (AKT)/Forkhead box O3 (FOXO3a) in type II alveolar epithelial cells [34], and myosin light chain kinase (MLCK) signaling pathways. Previous studies reported dietary Ile improved antioxidant status by regulating Nrf2 mRNA expression in intestine of Jian carp and increased TJs expressions in gill of grass carp [29,30]....

Show abstract

Bone marrow mesenchymal stem cell-derived exosomes alleviate hyperoxia-induced lung injury via the manipulation of microRNA-425

2021, Archives of Biochemistry and Biophysics

> *Citation Excerpt :*
>
> ...In addition, miR-425 in BMSCs-Exos activated the PI3K/AKT axis by targeting PTEN. Dias-Freitas et al. declare that signaling pathways transmission in HI cell injury was importantly associated with LI progression [33]. In hyperoxic condition, it was found that the generation of apoptotic cells could be induced by oxidative stress with the limitation of the PI3K/AKT axis [34]....

Show abstract

Hyperoxia exposure upregulates Dvl-1 and activates Wnt/β-catenin signaling pathway in newborn rat lung

2023, BMC Molecular and Cell Biology

The role and mechanism of miR-92a in endothelial autophagy

2023, Research Square

Melatonin Protects Against Hyperoxia-Induced Apoptosis in Alveolar Epithelial type IICells by Activating the MT2/PI3K/AKT/ETS1 Signaling Pathway

2023, Lung

View all citing articles on Scopus

Recommended articles (6)

Research article

Surfactant protein D regulates caspase-8-mediated cascade of the intrinsic pathway of apoptosis while promoting bleb formation

Molecular Immunology, Volume 92, 2017, pp. 190-198

Show abstract

Research article

Structural basis for substrate specificity of *meso*-diaminopimelic acid decarboxylase from *Corynebacterium glutamicum*

Biochemical and Biophysical Research Communications, Volume 495, Issue 2, 2018, pp. 1815-1821

Show abstract

Research article

Elevated *O*-GlcNAcylation stabilizes FOXM1 by its reduced degradation through GSK-3β inactivation in a human gastric carcinoma cell line, MKN45cells

Biochemical and Biophysical Research Communications, Volume 495, Issue 2, 2018, pp. 1681-1687

Show abstract

Research article

DANFIN functions as an inhibitor of transcription factor NF-κB and potentiates the antitumor effect of bortezomib in multiple myeloma

Biochemical and Biophysical Research Communications, Volume 495, Issue 3, 2018, pp. 2289-2295

Show abstract

Research article

Modulators of inflammation in Bronchopulmonary Dysplasia

Seminars in Perinatology, Volume 42, Issue 7, 2018, pp. 459-470

Show abstract

Research article

Hydrogen protects auditory hair cells from cisplatin-induced free radicals

Neuroscience Letters, Volume 579, 2014, pp. 125-129

Show abstract

View full text