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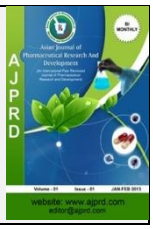

Open Access Research Article

Ginsenoside Rg3 Alleviates Rotenone-induced Lung Injury in Mice by Its Anti-oxidative Properties

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ABSTRACT

Objectives: Acute lung injury is commonly found in patients with insecticide poisoning and the pathogenesis is related to oxidative stress. Ginsenoside Rg3, one of the main constituents of *Panax ginseng C.A.* Meyer, shows an anti-oxidative activity. The aim of this study was to evaluate whether ginsenoside Rg3 can alleviate lung injury induced by rotenone in mice.
Methods: C57BL/6J male mice were divided into five groups (n=11). The mice in ginsenoside Rg3 groups were treated with ginsenoside Rg3 at dose of 5, 10 or 20 mg/kg. Except for the control group, mice were challenged intragastrically with rotenone at dose of 30 mg/kg, once a day for 6 weeks. Subsequently, the lung tissues of mice were collected. The effect of ginsenoside Rg3 on rotenone-induced lung injury was observed by hematoxylin and eosin staining. The oxidative stress in lung tissues were also examined.
Results: Rotenone induced substantial hemorrhage, alveolar wall thickness and neutrophils infiltration. These structural damages were attenuated significantly by ginsenoside Rg3 treatment. The lung injury induced by rotenone was associated with oxidative stress in lung tissues of mice. Compared with the control group, rotenone exposure resulted in the increase of malondialdehyde (MDA), the decreases of the activities of superoxide dismutase (SOD), glutathione peroxidase (GHS-Px), and the glutathione (GSH) content. Nevertheless, ginsenoside Rg3 treatment not only reduced MDA production but also increased the activities of SOD, GSH-Px, and the content of GSH in lung tissue of mice.
Conclusion: Taken together, this study demonstrated that ginsenoside Rg3 has potential to ameliorate rotenone-induced lung injury and the mechanism of action of ginsenoside Rg3 is mediated by its anti-oxidative properties.

Key words: Ginsenoside Rg3, rotenone, acute lung injury, oxidative stress

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INTRODUCTION

Insecticide abuse causes a serious pollution to the environment during agricultural activities. Moreover, the data of World Health Organization (WHO) show that the insecticide poisoning is the leading cause of suicide worldwide [(1)]. And insecticide poisoning induces more than 200,000 people's death every year [(2)]. This severe insecticide poisoning causes a damage to organs of human body. The symptoms of insecticide poisoning are coma, renal injury, acute lung injury (ALI), and respiratory failure [(3)]. ALI, initial state of acute respiratory distress syndrome (ARDS), presents mainly with acute respiratory insufficiency, which is implicated in alveolar epithelial injury and capillary endothelial cell injury. Patients with ALI/ARDS occupy for 10% in intensive care unit (ICU) and 4% in total inpatients. It is the main reason of mortality in critically ill patients. Generally, the mortality of patients with ALI is up to 60%, which is a major medical issue to be resolved so far [(4)].

Rotenone is used commonly as an insecticide in agriculture. It has the effect of inhibiting mitochondrial complex I which is an important metabolic enzyme involved in redox reactions [5]. The inhibition of mitochondrial complex I leads to mitochondrial dysfunction and trigger oxidative stress occurrence [6-7]. In animal experiments, many reports demonstrated that lung injury of rat were induced by an exposure of rotenone [8-9]. Previous study shows that oxidative stress is a key contributing factor for the lung injury and the impairment of lung function [10]. Meanwhile, reactive oxygen species-induced oxidative stress also plays an important role in the pathogenesis of respiratory diseases [11]. Recently, researches demonstrate that using antioxidants against oxidative stress reduces the airway inflammation in animal models of asthma [12]. Epigallocatechin-3-gallate (EGCG), a catechin in green tea, shows an antioxidant activity and therefore attenuates lung injury [13]. Thus, reducing oxidative stress may be an effective strategy to against insecticide poisoning-induced lung injury.

Ginsenosides are the compounds extracted from *Panax ginseng C.A.* Meyer. Up to date, more than 30 ginsenosides including Rg3, Rg2, Re, and Rh2 were separated [14]. Ginsenoside Rg3 is a tetracyclic triterpenoid saponin. Previous studies show that ginsenoside Rg3 possesses a good many of pharmacological activities, such as antioxidant [15], anticancer [16], cardiovascular protection [17], immune improvement [18], anti-inflammation [19], and anti-aging effects [20]. Ginsenoside Rg3 alleviates the dopaminergic nerve injury in Parkinson's disease mice through its antioxidant effects [21-22]. Ginsenoside Rg3 exerts an antioxidant activity and then ameliorates myocardial ischemia-reperfusion injury in rats [23]. This study aims to investigate whether ginsenoside Rg3 can ameliorate lung injury induced by rotenone in mice.

MATERIALS AND METHODS

Chemicals and Reagents

Rotenone was purchased from Sigma-Aldrich (St. Louis, MO, USA). Ginsenoside Rg3 was provided by Dalian Fusheng Natural Medicine Development Co., Ltd. (Dalian, China). Carboxymethycellulose sodium was purchased from Guangfu Fine Chemical Company (Tianjin, China). Malondialdehyde (MDA) kit (Lot. 20200508), superoxide dismutase (SOD) kit (Lot. 20200508), glutathione (GSH) kit (Lot. 20200416), and glutathione peroxidase (GHS-Px) kit (Lot. 20200416) were purchased from Nanjing Jiancheng Bioengineering Institute (Nanjing, China).

Animals

C57BL/6J male mice (18 - 22 g) were from Jinan Pengyue Experimental Animal Breeding Company (Jinan, China). The animals were housed in a specific pathogen-free conditions and temperature-controlled facility with a 12-hr light/dark cycle, and had *ad libitum* access to a standard animal diet. All procedures were performed in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals (Publication 86–23, revised in 1986), and the protocols were approved by the Ethics Committee of Yantai University (No. 58-ECYU-19).

Experimental Design

Animals were randomly divided into five groups (n=11) including the control, model, and ginsenoside Rg3 (5, 10, and 20 mg/kg) groups. Rotenone and ginsenoside Rg3 were dissolved in 0.5% carboxymethycellulose sodium. Mice in control and model groups were administered intragastrically with 0.5% carboxymethycellulose sodium. While mice in ginsenoside Rg3 groups were treated intragastrically with ginsenoside Rg3 at doses of 5, 10, or 20 mg/kg. Two hours later, mice in the control group were intragastrically with 0.5% carboxymethycellulose sodium again. Mice in the other groups were intragastrically challenged with rotenone (30 mg/kg) to induce the lung injury model. The treatment with ginsenoside Rg3 and the challenge with rotenone were performed once a day for 6 weeks.

Histomorphology

Mice were anesthetized with ketamine (90 mg/kg) and xylazine (5 mg/kg). The lung tissues were washed with 0.9% saline and then they were fixed with 4% paraformaldehyde. After dehydrated and embedded, the lung tissues were consecutively cut into 4-μm thick sections for hematoxylin and eosin (H&E) staining. The pathological changes were evaluated by three pathologists who were blinded to the design with an inverted microscope (IX71, Olympus, Japan). The lung injury was detected by Smith scoring criteria: pulmonary hyaline membrane formation, alveolar cavity enlargement, alveolar wall thickening, hemorrhage, necrosis, and inflammatory cell infiltration. The severity of lung injury was graded from 0 to 4.

Oxidative Stress Measurement

Oxidative stress indicators in the lung tissues of mice, such as MDA, SOD, GSH, and GHS-Px, were assayed according to the manufacturer's instructions. Briefly, the lung tissues from eight mice in each group were weighed and homogenized in phosphate-buffered saline at a ratio of 1:10 (weight to volume). The homogenates were centrifuged at 16, 000 *g* at 4 ℃ for 10 min. The supernatant was used to measure oxidative stress indicators. Data were expressed as the absorbance after normalization by the protein content.

Statistical Analysis

Data are presented as the Mean ± SD. Experimental data were analyzed by SPSS 19.0 statistical analysis (IBM Corporation, Armonk, NY, USA). Differences among multiple groups were analyzed by One-way ANOVA followed by Fisher's LSD test. A p-value < 0.05 was considered to indicate a significant difference.

RESULTS

The effect of ginsenoside Rg3 on rotenone-induced lung injury in mice

As shown in Fig. 1, the pathological section of lung tissues in the control group demonstrated that the bronchial epithelial structure was complete, the structure of epithelial cells was normal and arranged tightly, and the alveolar structure was clear. Furthermore, there was no obvious thickness of the alveolar wall and inflammation in the control group. However, substantial hemorrhage, alveolar wall thickness, and neutrophils infiltration were observed in

the model group. Treatment with ginsenoside Rg3 alleviated the rotenone-induced lung injury. Especially, there were only slight alveolar epithelial cell hyperplasia and alveolar septal thickness in the lungs of the mice of ginsenoside Rg3 at dose of 20 mg/kg group. The score of lung injury was shown in Fig. 2. Compared with the control group, the score of lung injury in the model group increased significantly (p<0.01). However, treatment with ginsenoside Rg3 at dose of 5, 10, and 20 mg/kg decreased the score of lung injury when compared with that of the model group (p<0.01). These results demonstrated that ginsenoside Rg3 had a property to alleviate the lung injury induced by rotenone in mice.

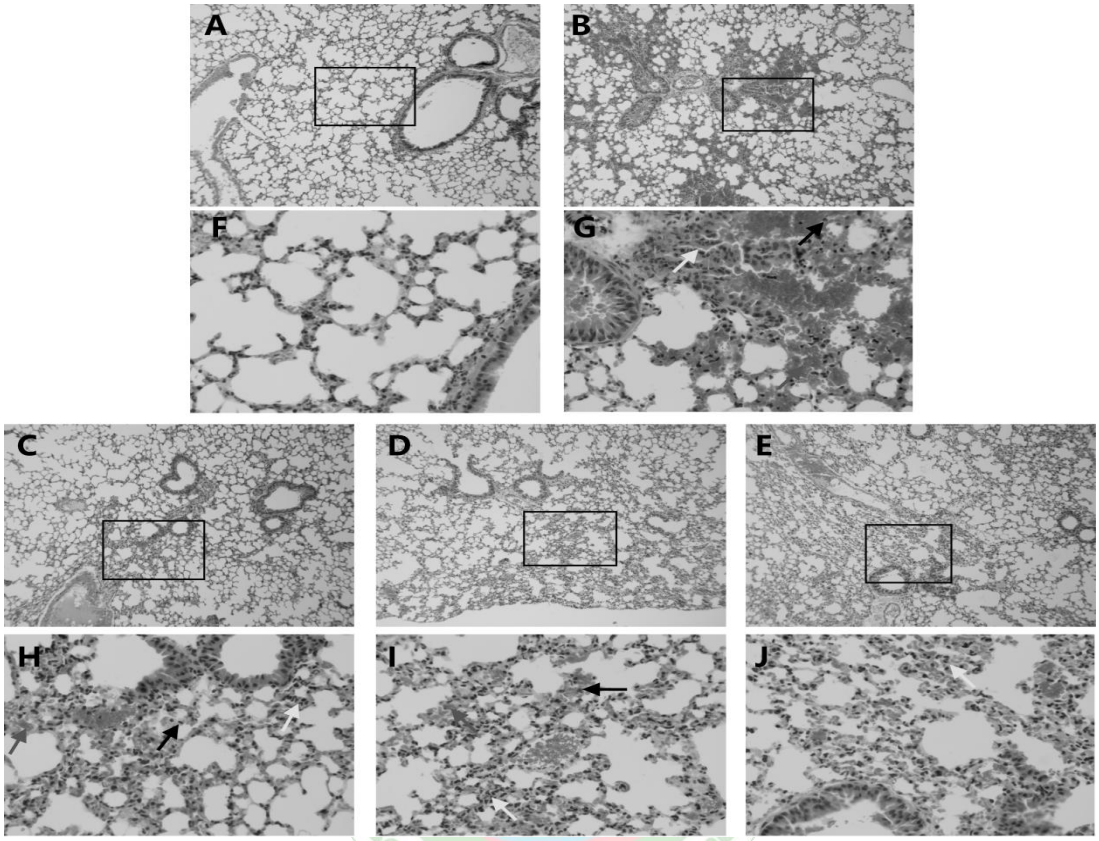

Fig. 1. The effect of ginsenoside Rg3 on rotenone-induced lung injury in mice. Lung histopathology of mice stained with H&E (n=3). A, Control; B, Model; C, Ginsenoside Rg3 5 mg/kg; D, Ginsenoside Rg3 10 mg/kg; E, Ginsenoside Rg3 20 mg/kg, (×100). F, Control; G, Model; H, Ginsenoside Rg3 5 mg/kg; I, Ginsenoside Rg3 10 mg/kg; J, Ginsenoside Rg3 20 mg/kg, (×400). Red arrowheads indicate hemorrhage, yellow arrowheads indicate alveolar wall thickening and alveolar epithelial cell hyperplasia, black arrowheads indicate inflammatory cell infiltration.

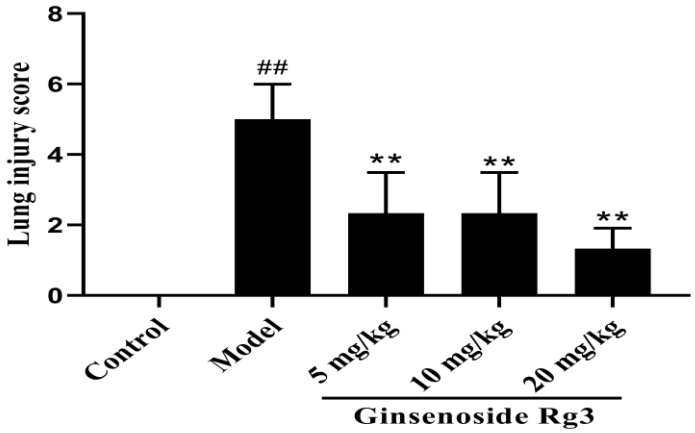

Fig. 2. The effect of ginsenoside Rg3 on rotenone-induced lung injury in mice. The score of lung injury. Data were expressed as the Mean ± SD (n = 3). Statistical analyses were performed using One-way ANOVA followed by Fisher's LSD test. ##p<0.01 compared with the control group; **p<0.01compared with the model group.

The effect of ginsenoside Rg3 on SOD activity in the lung tissues

SOD is an important antioxidant enzyme and presents a property to against oxidative stress. As shown in Fig. 3, the SOD activity of model group was lower than that of the control group (p<0.01). However, treatment with

ginsenoside Rg3 at dose of 10 and 20 mg/kg markedly increased the SOD activity in the lung tissues of mice (p<0.05 or 0.01).

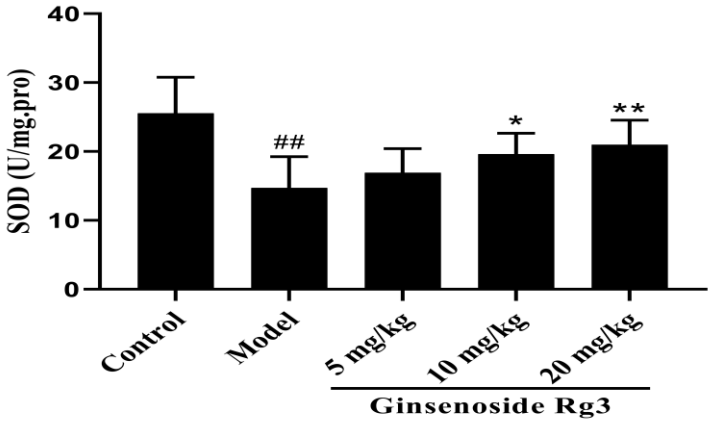

Fig. 3. The effect of ginsenoside Rg3 on SOD activity in the lung tissues. Bar graphs of SOD activity in lung tissue. Data were expressed as the Mean ± SD (n = 8). Statistical analyses were performed using One-way ANOVA followed by Fisher's LSD test. ##p<0.01 compared with the control group; *p<0.05, **p<0.01 compared with the model group.

The effect of ginsenoside Rg3 on GSH-Px activity and GSH content in the lung tissues

In Fig. 4, GSH-Px activity of the model group decreased significantly when compared with the control group (p<0.01). Ginsenoside Rg3 augmented the GSH-Px activity (p< 0.05 or p< 0.01). Consistently, GSH in the lung tissues of model group was also decreased when compared with the control group (p<0.01, Fig.5). Treatment with ginsenoside Rg3 at dose of 5, 10 and 20 mg/kg increased the GSH content in the lung tissues (p<0.05 or p< 0.01).

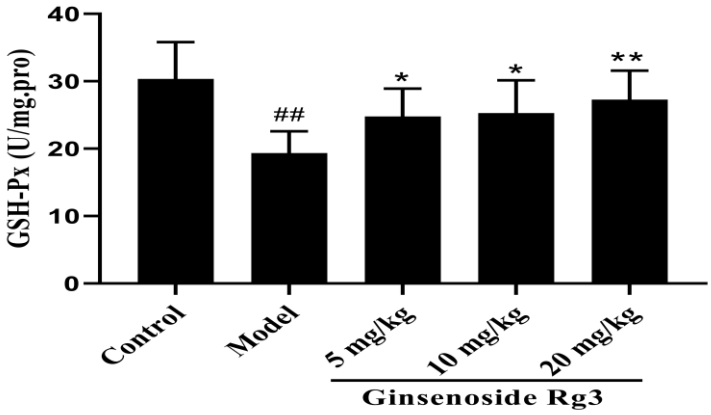

Fig. 4. The effect of ginsenoside Rg3 on GSH-Px activity in the lung tissues. Bar graphs of GSH-Px activity in lung tissue. Data were expressed as the Mean ± SD (n = 8). Statistical analyses were performed using One-way ANOVA followed by Fisher's LSD test. ##p<0.01 compared with the control group; *p<0.05, **p<0.01 compared with the model group.

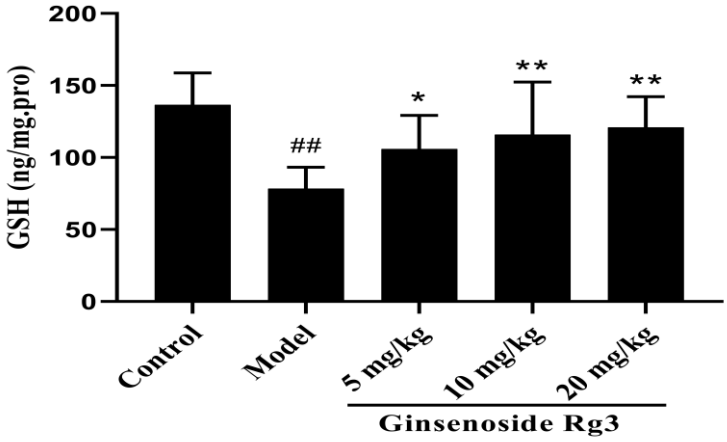

Fig. 5. The effect of ginsenoside Rg3 on GSH content in the lung tissues. Bar graphs of GSH content in lung tissue. Data were expressed as the Mean ± SD (n = 8). Statistical analyses were performed using One-way ANOVA followed by Fisher's LSD test. ##p<0.01 compared with the control group; *p<0.05, **p<0.01 compared with the model group.

The effect of ginsenoside Rg3 on MDA content in the lung tissues

MDA is a parameter reflecting the degree of lipid peroxidation. As shown in Fig. 6, there was an increase of MDA in the lung tissues of the mice of the model group (p<0.01). Compared with the model group, the MDA content in ginsenoside Rg3 groups was decreased significantly (p<0.01). The results indicated that ginsenoside Rg3 alleviates rotenone-induced lung injury in mice by its anti-oxidative properties.

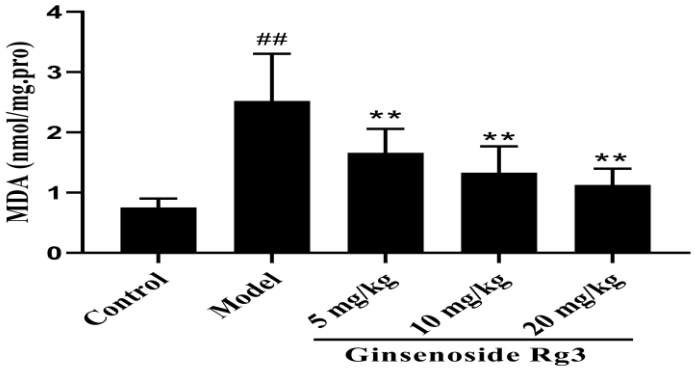

Fig. 6. The effect of ginsenoside Rg3 on MDA content in the lung tissues. Bar graphs of MDA content in lung tissue. Data were expressed as the Mean ± SD (n = 8). Statistical analyses were performed using One-way ANOVA followed by Fisher's LSD test. ##p<0.01 compared with the control group; **p<0.01 compared with the model group.

DISCUSSION

In this study, rotenone exposure caused a severe lung injury and an oxidative stress of lung in mice. However, ginsenoside Rg3 alleviated the lung injury induced by rotenone. Further experiments demonstrated that the mechanism of action of ginsenoside Rg3 is mediated by its anti-oxidative properties.

Insecticides, such as rotenone, omethoate, methyl parathion and dichlorvos, can cause oxidative stress and severe lung damage [24-26]. Researches have demonstrated that exposure of insecticides for long time caused pulmonary impairments in mice and rabbits [27]. Besides, oxidative stress and inflammation in lung were major features in the insecticides-challenged rats [28]. Rotenone is mainly extracted from the roots and stems of *Lonchocarpus* and *Derris* species [29]. It is a commonly used insecticide in agriculture. Previous studies indicated that rotenone could cause a mitochondrial dysfunction, which is implicated in the inhibition of mitochondrial electron transport chain complex I [30]. Therefore, the leak of electrons from mitochondria will react with oxygen and produce reactive oxygen species (ROS). ROS leads to the mitochondrial disorders, abnormal nucleic acids, protein misfolding, and disorders of lipid metabolism. Previous study showed that rotenone caused the injury of lung in mouse models [31]. In this study, histopathology evaluation was performed to determine the lung injury in the rotenone-challenged mice. The results showed that rotenone exposure induced a severe damage in lung tissue of mice, including hemorrhage and alveolar wall thickness. However, ginsenoside Rg3 treatment alleviated rotenone-induced lung injury in mice. These findings demonstrated that ginsenoside Rg3 can alleviates rotenone-induced lung injury in mice.

The molecular mechanisms of insecticides-induced lung injury are complex and diverse. But the main pathological mechanisms include inflammation, oxidative stress, and apoptosis [28,32]. Accordingly, rotenone-induced lung injury was implicated in the increase of ROS under the condition of oxidative stress [4-5] Although lung has its antioxidant system, excessive ROS will result in the damage of epithelial cells of lung [33-34]. Moreover, clinical studies also demonstrated that ROS plays a vital role in airway tissue damage of patients [4]. Emerging evidence suggests that oxidative stress was one of the main factors which causes lung injury [35-37]. Thus, oxidative stress plays an indispensable role in the occurrence and development of lung injury induced by rotenone. Therefore, reducing oxidative stress and/or increasing antioxidant capacity are primary treatment strategies for alleviating rotenone-induced lung injury. Cells can also prevent oxidative stress-induced damage by increasing the activities of the antioxidant enzymes [38]. The antioxidant enzymes, including GSH-Px and SOD, can scavenge ROS and therefore prevent the damage of oxidative stress [39]. GSH is a cardinal antioxidant in cells and it protects cells against exogenous and endogenous toxins, including ROS and nitrogen species. Previous study showed that ovalbumin inhalation increased the ROS generation in bronchoalveolar lavage fluids. But the augment of GSH content significantly reduced the lung injury induced by the ovalbumin inhalation [40]. MDA is a stable product of lipid peroxidation of cells and therefore is the most frequently used biomarker of oxidative stress. In this study, rotenone exposure resulted in the increase of MDA, the decreases of the activities of SOD, GSH-Px, and the GSH content. Nevertheless, ginsenoside Rg3 treatment not only reduced MDA production but also increased the activities of SOD, GSH-Px, and the content of GSH in lung tissue of mice. The findings of these experiments demonstrated that the mechanism of action of ginsenoside Rg3 is mediated by its anti-oxidative properties.

There are several limitations in this study. ROS is an important indicator of insecticides-caused lung injury. However, the levels of ROS were not assayed directly in the present study. Additionally, lactate acid level and lactate clearance show a direct relationship with mortality in patients with lung injury in clinic [41]. Therefore, it will be better to detect the lactate acid content in the blood of mice with rotenone exposure.

CONCLUSION

In summary, this study demonstrates that ginsenoside Rg3 exerts lung protective effects in rotenone-induced lung injury mice. And the mechanism of action of ginsenoside Rg3 is mediated by its anti-oxidative properties.

ACKNOWLEDGMENTS

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REFERENCES

1. Bertolote JM, Fleischmann A, Butchart A, Besbelli N. Suicide, suicide attempts and pesticides: a major hidden public health problem. Bulletin of the World Health Organization. 2006; 84(4):260. doi: 10.2471/blt.06.030668.
2. Gunnell D, Eddleston M, Phillips MR, Konradsen F. The global distribution of fatal pesticide self-poisoning: systematic review. BioMed Central public health. 2007; 7:357. doi: 10.1186/1471-2458-7-357.
3. Hulse EJ, Clutton RE, Drummond G, Eddleston M. Translational toxicological research: investigating and preventing acute lung injury in organophosphorus insecticide poisoning. Journal of the Royal Army Medical Corps. 2014; 160(2):191-2. doi: 10.1136/jramc-2013-000207.
4. Bellani G, Laffey JG, Pham T, Fan E, Brochard L et al. Epidemiology, Patterns of Care, and Mortality for Patients With Acute Respiratory Distress Syndrome in Intensive Care Units in 50 Countries. Journal of the American Medical Association. 2016; 315(8):788-800. doi: 10.1001/jama.2016.0291.
5. Li N, Ragheb K, Lawler G, Sturgis J, Rajwa B et al. Mitochondrial complex I inhibitor rotenone induces apoptosis through enhancing mitochondrial reactive oxygen species production. The Journal of biological chemistry. 2003; 278(10):8516-25. doi: 10.1074/jbc.M210432200.
6. Hasan W, Kori RK, Jain J, Yadav RS, Jat D. Neuroprotective effects of mitochondria-targeted curcumin against rotenone-induced oxidative damage in cerebellum of mice. Journal of biochemical and molecular toxicology. 2020; 34(1):e22416. doi: 10.1002/jbt.22416.
7. Yu JH, Huang CF, Wang TH, Hung DZ, Mu HW et al. Oxidative storm in a patient with acute rotenone-containing plant poisoning. The American journal of emergency medicine. 2020; 38(6):1296. doi: 10.1016/j.ajem.2020.01.019.
8. Bongard RD, Yan K, Hoffmann RG, Audi SH, Zhang X et al. Depleted energy charge and increased pulmonary endothelial permeability induced by mitochondrial complex I inhibition are mitigated by coenzyme Q1 in the isolated perfused rat lung. Free radical biology & medicine. 2013; 65:1455-1463. doi: 10.1016/j.freeradbiomed.2013.07.040.
9. 9.Audi SH, Merker MP, Krenz GS, Ahuja T, Roerig DL et al. Coenzyme Q1 redox metabolism during passage through the rat pulmonary circulation and the effect of hyperoxia. Journal of applied physiology (Bethesda, Md. : 1985). 2008; 105(4):1114-26. doi: 10.1152/japplphysiol.00177.2008.
10. Galam L, Failla A, Soundararajan R, Lockey RF, Kolliputi N. 4-hydroxynonenal regulates mitochondrial function in human small airway epithelial cells. Oncotarget. 2015; 6(39):41508-21. doi: 10.18632/oncotarget.6131.
11. Dworski R. Oxidant stress in asthma. Thorax. 2000; 55 Suppl 2(Suppl 2):S51-3. doi: 10.1136/thorax.55.suppl_2.s51.
12. Cho YS, Lee J, Lee TH, Lee EY, Lee KU et al. Alpha-Lipoic acid inhibits airway inflammation and hyperresponsiveness in a mouse model of asthma. The Journal of allergy and clinical immunology. 2004; 114(2):429-35. doi: 10.1016/j.jaci.2004.04.004.
13. Luo ZL, Sun HY, Wu XB, Cheng L, Ren JD. Epigallocatechin-3-gallate attenuates acute pancreatitis induced lung injury by targeting mitochondrial reactive oxygen species triggered NLRP3 inflammasome activation. Food & function. 2021; 12(12):5658-5667. doi: 10.1039/d1fo01154e.
14. Park SK, Hyun SH, In G, Park CK, Kwak YS et al. The antioxidant activities of Korean Red Ginseng (Panax ginseng) and ginsenosides: A systemic review through in vivo and clinical trials. Journal of ginseng research. 2021; 45(1):41-47. doi: 10.1016/j.jgr.2020.09.006.
15. Wei X, Su F, Su X, Hu T, Hu S. Stereospecific antioxidant effects of ginsenoside Rg3 on oxidative stress induced by cyclophosphamide in mice. Fitoterapia. 2012; 83(4):636-42. doi: 10.1016/j.fitote.2012.01.006.
16. Sun M, Ye Y, Xiao L, Duan X, Zhang Y et al. Anticancer effects of ginsenoside Rg3 (Review). International journal of molecular medicine. 2017; 39(3):507-518. doi: 10.3892/ijmm.2017.2857.
17. Lee H, Kong G, Tran Q, Kim C, Park J et al. Relationship Between Ginsenoside Rg3 and Metabolic Syndrome. Frontiers in pharmacology. 2020; 11:130. doi: 10.3389/fphar.2020.00130.
18. Liu X, Zhang Z, Liu J, Wang Y, Zhou Q et al. Ginsenoside Rg3 improves cyclophosphamide-induced immunocompetence in Balb/c mice. International immunopharmacology. 2019; 72:98-111. doi: 10.1016/j.intimp.2019.04.003.
19. Yang J, Li S, Wang L, Du F, Zhou X et al. Ginsenoside Rg3 Attenuates Lipopolysaccharide-Induced Acute Lung Injury via MerTK-Dependent Activation of the PI3K/AKT/mTOR Pathway. Frontiers in pharmacology. 2018; 9:850. doi: 10.3389/fphar.2018.00850.
20. Lee H, Hong Y, Tran Q, Cho H, Kim M et al. A new role for the ginsenoside RG3 in antiaging via mitochondria function in ultraviolet-irradiated human dermal fibroblasts. Journal of ginseng research. 2019; 43(3):431-441. doi: 10.1016/j.jgr.2018.07.003.
21. Han Y, Wang T, Li C, Wang Z, Zhao Y et al. Ginsenoside Rg3 exerts a neuroprotective effect in rotenone-induced Parkinson's disease mice via its anti-oxidative properties. European journal of pharmacology. 2021; 909:174413. doi: 10.1016/j.ejphar.2021.174413.
22. Cheng Z, Zhang M, Ling C, Zhu Y, Ren H et al. Neuroprotective Effects of Ginsenosides against Cerebral Ischemia. Molecules (Basel, Switzerland). 2019; 24(6):1102. doi: 10.3390/molecules24061102.
23. Li L, Wang Y, Guo R, Li S, Ni J et al. Ginsenoside Rg3-loaded, reactive oxygen species-responsive polymeric nanoparticles for alleviating myocardial ischemia-reperfusion injury. Journal of controlled release: official journal of the Controlled Release Society. 2020; 317:259-272.doi: 10.1016/j.jconrel.2019.11.032.
24. He F, Wang J, Liu Y, Wang X, Cai N et al. Xuebijing injection induces anti-inflammatory-like effects and downregulates the expression of TLR4 and NF-κB in lung injury caused by dichlorvos poisoning. Biomedicine & pharmacotherapy. 2018; 106:1404-1411. doi: 10.1016/j.biopha.2018.07.111.
25. Pinton S, Luchese C, Stangherlin EC, Nogueira CW. Acute exposure to diphenyl ditelluride causes oxidative damage in rat lungs. Ecotoxicology and environmental safety. 2011; 74(3):521-6. doi: 10.1016/j.ecoenv.2010.07.031.
26. Wang J, Yu XF, Zhao JJ, Shi SM, Fu L et al. Ginsenoside Rg3 attenuated omethoate-induced lung injury in rats. Human & experimental toxicology. 2016; 35(6):677-84. doi: 10.1177/0960327115597984.
27. Yavuz Y, Kaya E, Yurumez Y, Sahin O, Bas O et al. Technetium-99m diethylenetriaminepentaacetic acid radioaerosol scintigraphy in organophosphate induced pulmonary toxicity: experimental study. Clinical toxicology (Philadelphia, Pa.). 2008; 46(8):711-5. doi:

10.1080/15563650701584111.

28. Du Y, Wang T, Jiang N, Ren RT, Zhao DL et al. Protective effect of sodium aescinate on lung injury induced by methyl parathion. Human & experimental toxicology. 2011; 30(10):1584-91. doi: 10.1177/0960327110393764.
29. Ramalingam M, Huh YJ, Lee YI. The Impairments of α-Synuclein and Mechanistic Target of Rapamycin in Rotenone-Induced SH-SY5Y Cells and Mice Model of Parkinson's Disease. Frontiers in neuroscience. 2019; 13:1028. doi: 10.3389/fnins.2019.01028.
30. Tanner CM, Kamel F, Ross GW, Hoppin JA, Goldman SM et al. Rotenone, paraquat, and Parkinson's disease. Environmental health perspectives. 2011; 119(6):866-72. doi: 10.1289/ehp.1002839.
31. Ahmad T, Mukherjee S, Pattnaik B, Kumar M, Singh S et al. Miro1 regulates intercellular mitochondrial transport & enhances mesenchymal stem cell rescue efficacy. The European Molecular Biology Organization journal. 2014; 33(9):994-1010. doi: 10.1002/embj.201386030.
32. Shaikh NI, Sethi RS. Impairment of apoptosis pathway via Apaf1 downregulation during chlorpyrifos and/or cypermethrin induced lung damage. Animal Biotechnology. 2021:1-8. doi: 10.1080/10495398.2021.1981918.
33. Rahman I, Morrison D, Donaldson K, MacNee W. Systemic oxidative stress in asthma, COPD, and smokers. American journal of respiratory and critical care medicine. 1996; 154(4 Pt 1):1055-60. doi: 10.1164/ajrccm.154.4.8887607.
34. Cortijo J, Martí-Cabrera M, de la Asuncíon JG, Pallardó FV, Esteras A et al. Contraction of human airways by oxidative stress protection by N-acetylcysteine. Free radical biology & medicine. 1999; 27(3-4):392-400. doi: 10.1016/s0891-5849(99)00070-2.
35. Yang H, Lv H, Li H, Ci X, Peng L. Oridonin protects LPS-induced acute lung injury by modulating Nrf2-mediated oxidative stress and Nrf2-independent NLRP3 and NF-κB pathways. Cell communication and signaling: CCS. 2019; 17(1):62. doi: 10.1186/s12964-019-0366-y.
36. Li T, Wu YN, Wang H, Ma JY, Zhai SS et al. Dapk1 improves inflammation, oxidative stress and autophagy in LPS-induced acute lung injury via p38MAPK/NF-κB signaling pathway. Molecular immunology. 2020; 120:13-22. doi: 10.1016/j.molimm.2020.01.014.
37. Jiang JS, Chou HC, Chen CM. Cathelicidin attenuates hyperoxia-induced lung injury by inhibiting oxidative stress in newborn rats. Free radical biology & medicine. 2020; 150:23-29. doi: 10.1016/j.freeradbiomed.2020.02.005.
38. Jenner P. Oxidative stress in Parkinson's disease. Annals of neurology. 2003;53 Suppl 3:S26-36; discussion S36-8. doi: 10.1002/ana.10483.
39. Zhang H, Zheng W, Feng X, Yang F, Qin H et al. Nrf2-ARE Signaling Acts as Master Pathway for the Cellular Antioxidant Activity of Fisetin. Molecules (Basel, Switzerland). 2019; 24(4):708. doi: 10.3390/molecules24040708.
40. Lee YC, Lee KS, Park SJ, Park HS, Lim JS et al. Blockade of airway hyperresponsiveness and inflammation in a murine model of asthma by a prodrug of cysteine, L-2-oxothiazolidine-4-carboxylic acid. FASEB journal: official publication of the Federation of American Societies for Experimental Biology. 2004; 18(15):1917-9. doi: 10.1096/fj.04-2212fje.
41. Ryoo SM, Lee J, Lee YS, Lee JH, Lim KS et al. Lactate Level Versus Lactate Clearance for Predicting Mortality in Patients With Septic Shock Defined by Sepsis-3. Critical care medicine. 2018; 46(6):e489-e495. doi: 10.1097/CCM.0000000000003030.