PIBB 生物化学与生物物理进展
Progress in Biochemistry and Biophysics
2020,47(8):762~767
www.pibb.ac.cn

Dragon's Blood Has Protective Effects in Irradiation-induced Neuroinflammation*

ZHANG Peng[1)**], WANG Hai-Long[1,2)**], MA Hong[1)], SHEN Qian-Wei[1)], SUN Fei-Yi[1)], CHEN Yu[1)], LAN Yu[1)], DAI Rong-Ji[1)], DENG Yu-Lin[1)***]

([1)]*School of Life Science, Beijing Institute of Technology, Beijing* 100081, China;
[2)]*Laborary of Cancer Cell Biology, Tianjin Medical University Cancer Institute& Hospital, Tianjin* 300060, China)

Abstract The continuous development of deep space exploration, the potential threat of war, and the occurrence of nuclear accidents increase our exposure to irradiation. Irradiation-induced neuroinflammation is one of the important aspects. The occurrence of neuroinflammation is closely related to neurodegenerative diseases such as Alzheimer's disease (AD) and Parkinson's disease (PD). The development of traditional Chinese medicines (TCM) with antioxidant and anti-inflammatory effects has positive implications for irradiation-induced damage to the central nervous system (CNS). Traditional Chinese medicine dragon's blood has a good effect on the treatment of radiation-induced neuroinflammation. This review summarizes the role of dragon's blood in reducing oxidative stress levels, expression of related inflammatory factors, and mitochondrial damage. At the same time, we propose that endogenous neurotoxins may aggravate brain-induced neuroinflammation which might be alleviated by dragon's blood.

Key words dragon's blood, irradiation, ROS, neuroinflammation, mitochondrial, endogenous neurotoxin
DOI: 10.16476/j.pibb.2020.0246

During long-term space travel, astronauts are easily exposed to the Milky Way Cosmic Ray (GCR) and Solar Particle Events[1], which poses a great health risk to astronauts. Among these injuries, damage to the CNS may cause a series of problems, including learning, memory, processing speed, attention, cognitive flexibility, and executive functions like cranial radiotherapy to prevent malignant progression of the brain[2]. Studies have shown that in the three-year Mars mission, it is estimated that 13% of neurons in the CNS will be infiltrated by iron ions at least once, while about 50% of neurons in hippocampal neurons are infiltrated[3]. In addition, due to the proliferation of nuclear weapons, potential terrorist attacks, natural disasters at nuclear power plan, and the widespread use of nuclear medicine, the risk of exposure to debilitating and lethal doses of irradiation is increasing for ordinary people[4]. Irradiation-induced CNS damage is often accompanied by an increase in Reactive oxygen species (ROS), which is a key signaling molecule in the inflammatory response[5].

TCM, as one of the most important parts of complementary and alternative medicine (CAM), plays a key role in the formation of integrated Chinese and Western medicine[6]. In the context of the increasing number of alternative forms of health care such as TCM in many Western countries, countries such as Australia, Canada and the United Kingdom have even begun to develop regulations, education and standards on CM practices[7]. The use of Dragon's Blood can be traced back to ancient Greece and the ancient Arab era, and its use as a TCM in China has also exceeded 1 500 years[8-10]. Li Shi-Zhen also recorded the medicinal value of the dragon's blood in Compendium of Materia Medica (Ming Dynasty, 1 368−1 644). Studies have shown that it has good effects in the treatment of blood stasis syndrome, anti-

* This work was supported by a grant from the Space Medical Experiment Project of China Manned Space Program (HYZHXM02003).
** These authors contributed equally to this work.
*** Corresponding author.
Tel: 86-10-68915996, E-mail: deng@bit.edu.cn
Received: July 11, 2020 Accepted: July 21, 2020

diarrhea, anti-ulcer, anti-bacterial, anti-viral, wound healing, anti-tumor, anti-inflammatory, anti-oxidation, improve blood circulation, metabolism and immune function[9,11].

The development of TCM with antioxidants and anti-inflammatory effects has a positive effect on the central nervous system risks in response to space and ground irradiation accidents. The present review enlightens Dragon's Blood is a positive effect in relieving irradiation-induced neuroinflammation.

1 Dragon's blood relates to anti-neuroinflammation after irradiation

1.1 Decrease the level of oxidative stress

Oxidative stress refers to an imbalance between oxidation and anti-oxidation in the body, which tends to oxidize, leading to stress, protein and DNA damage[12]. Chemically active oxygenated metabolites such as superoxide anion (O^{2-}), hydrogen peroxide (H_2O_2), and hydroxyl radicals (-OH) are produced by ROS during aerobic metabolism. Since the brain is the largest aerobic organ in the human body and contains oxidizable substances such as dopamine and lipids, the brain will cause an increase in oxidative stress after being exposed to irradiation. Irmak *et al*. [13] found that superoxide dismutase (SOD) and glutathione peroxidase (GSH-Px) caused 30 min or 60 min of irradiation by studying the level of oxidative stress in the fetal rat brain. The content decreased and the malondialdehyde (MDA) content increased. Moreover, low linear energy transfer (LET) γ rays and high LET carbon heavy ions also caused an increase in MDA and H_2O_2 content in the brain, a decrease in GSH, and a decrease in SOD and GSH-Px activity[9,14]. The anti-oxidation effect of dragon's blood on irradiation is reflected in various parts of the body. After the mice were subjected to γ ray whole body irradiation, the SOD, catalase (CAT) and GSH levels in the liver and spleen of the groups of Dragon's Blood and Dragon's Blood extract groups were significantly increased[15]. In addition, the level of MDA in serum is significantly reduced [16]. For the whole brain irradiation model, the dragon's blood protection group effectively increased the levels of SOD and GSH while reducing the levels of MDA and H_2O_2 in the brain[9]. In the rat model of cerebral ischemia, dragon's blood dropping pills can significantly increase the activity of GSH-PX and SOD, reduce the level of MDA, decrease the excess free radicals in rats, and induce the antioxidant activity of lipid peroxidation[17].

1.2 Inhibit the mitochondrial dysfunction

Mitochondria are a major natural cellular source of ROS, Kim *et al*. [18] showed that mitochondrial abnormalities are shown in unstable cells, and they contribute to oxidative stress in irradiation-induced genomic instability (RIGI) cells. After irradiation, the rapid production of -OH may oxidize biomolecules such as proteins or lipids in cells. These phospholipids are located in the mitochondrial inner membrane and are required for optimal activity of the electron transport chain complex, including complexes I and III. Therefore, shortly after irradiation, a strong release of -OH may damage mitochondria by peroxidation of these phospholipids within a few hours[19]. Loureirin B extracted from Dragon's Blood could inhibit ER stress-induced mitochondrial dysfunction and activate the Akt/GSK-3β pathway, which has a positive effect on axonal regeneration and motor function recovery[20].

1.3 Reduce the levels of inflammatory effects

Greene-Schloesser *et al*. [21] summarize changes in levels of related inflammatory factors in the brain after irradiation including: a. Upregulation of MCP-1/CCL2 and MIP-2/CXCL2 mRNA levels; b. Increased expression of pro-inflammatory molecules such as TNF-α, IL-1β, ICAM-1 and Cox-2; c. Activation pro-inflammatory transcription factors such as NF-κB; d. Acute infiltration of neutrophils and delayed increase in T cells, MHC II positive cells, and CD-11c positive cells. The levels of transcription of inflammatory cytokines, activated microglia and activated endothelial cells are strongly dependent on the metering and timing of irradiation[22]. Activated immune cells should be regulated to protect neurons from damage, but over-activated immune cells are toxic and it can be observed that irradiation-activated microglia can cause immune cells to infiltrate into the brain tissues, and then produce ROS, which in turn activates more microglia and induces more neural cells to maintain or increase oxidative stress levels. These findings indicate that the positive effects of traditional Chinese medicines with anti-inflammatory drugs and antioxidants would play roles in preventing or treating irradiation-induced neuroinflammation. Xin *et al*.[9] observed under the whole brain irradiation

model that dragon's blood can significantly reduce the levels of inflammatory cytokines such as TNF-α, INF-γ and IL-6 levels induced by irradiation in the brain tissues. Choy *et al*. [23] used dragon's blood ethanol extract to effectively inhibit endotoxin-induced NO, PGE 2, IL-1β and TNF-α release, as well as iNOS, p65 and COX-2 expression.

2 Drgon's blood promotes neuronal cells regeneration

As an inhibition of apoptosis and regeneration of neuronal cells, Brain Derived Neurotropic Factor (BDNF) is abundant in the hippocampus and plays an important role in learning and memory. Irradiation can significantly decrease the level of BDNF expression in the mouse hippocampus[24]. This leads to increased activations of Caspase proteins and memory deficits[24-25]. Our previous research had shown that Dragon's Blood could significantly increase the expression of BDNF levels and reduce caspase 3 levels, at 24 h, 3 d and 7 d compared with gamma irradiation groups[9].

3 The possible molecular mechanism of Dragon's Blood as an irradiation protectant

The protection effect of Dragon's Blood from brain damage under irradiation conditions mainly comes from its control of oxidative stress level and inflammation level[9]. Under non-stimulated conditions, NF-κB is masked by the inhibitory protein IκB-α, so the inactivated NF-κB cannot be transferred to the nucleus. However, NF-κB is transferred to the nucleus under the stimulation of activators such as IL-1β, TNF-α or cytokines. The mechanism is that NF-κB phosphorylates its inhibitory protein IκB-α, and then targets IκB-α for ubiquitination and degradation[26]. Choy *et al*. [23] found that cytosolic IκB-α elevation and nuclear NF-κB decreased in a lipopolysaccharide-stimulated RAW 264.7 cell treated with Dragon's Blood Extract (SDEE). This result indicated that SDEE may prevent inflammation by inhibiting NF-κB-mediated inflammatory genes such as iPS and COX-2. Li *et al*. [27] found that dragon's blood can block the synthesis and release of P (a well-known tachykinin peptide) by inhibiting COX-2 protein induction and intracellular calcium concentration, thereby exerting anti-inflammatory and analgesic effects.

Our previous research work described the oxidative stress, mitochondrial dysfunction, inflammation, alpha-synuclein aggregation abnormalities and endogenous neurotoxins involved in the pathogenesis of Parkinson's disease (PD), and explained the intrinsic relationship of these factors[28-29]. Briefly, this is a destructive feedback loop that includes three cycles, including: a. Oxidative stress aldehydes produced by lipid peroxidation can react with dopamine to form endogenous neurotoxins and may avoid oxide scavenging mechanisms; b. Continuously produced aldehydes, endogenous neurotoxins and/or damaged mitochondrial DNA, passively released from the intracellular space of damaged cells into the extracellular space, thereby inducing microglia and astrocytes activation, which enhances T cell infiltration, causes inflammation to exacerbate oxidative stress, thereby permanently forming endogenous neurotoxins; and c. Activation of microglia and astrocytes by inflammation greatly reduces their ability to clear abnormal alpha-synuclein. Abnormal alpha-synuclein inclusion bodies act as antigens to further induce inflammation and exacerbate oxidative stress levels, which again promote the formation of endogenous neurotoxins [28]. This causes us to think about the situation of radiation-induced brain damage and related neuroinflammation. Oxidative stress, inflammatory reaction and mitochondrial damage in the brain after irradiation damage may promote the occurrence of neuroinflammation, and there is a mutually reinforcing process. There have been few reports on radiation-induced endogenous neurotoxins, and we can be certain that endogenous neurotoxin formation may not occur preferentially after irradiation damage to the brain tissues, however, endogenous neurotoxins are most likely involved in the long-term effects of radiation-induced nerve injury or inflammation. Therefore, whether endogenous neurotoxins participate in this injury process requires further research.

4 Conclusion

The increase in irradiation leakage exacerbates the possibility of neuroinflammation. As we all know neuroinflammation plays an important role in the pathogenesis of various neurodegenerative diseases including Parkinson's disease and Alzheimer's disease.

Although there is no causal relationship between radiation and neurodegenerative diseases, receiving high doses of ionizing radiation on the head can lead to a series of neural inflammatory effects and behavioral changes including loss of pleasure, decreased desire to explore, and learning and memory impairment. Therefore, the development of anti-inflammatory and anti-oxidant traditional Chinese medicine plays an important role in preventing radiation-induced neuroinflammation. Dragon's blood is one of them, and a large number of studies have shown that it works well in the treatment of inflammation (Figure 1). However, at present, the research on irradiation protection of dragon's blood is mainly based on phenomena, and its mechanism of action has not been deeply studied. Therefore, we should focus on the research on dragon's blood protection mechanism to provide strong conditions for more efficient drug screening. At the same time, the role of endogenous neurotoxins in radiation-induced neuroinflammation should also be fully studied.

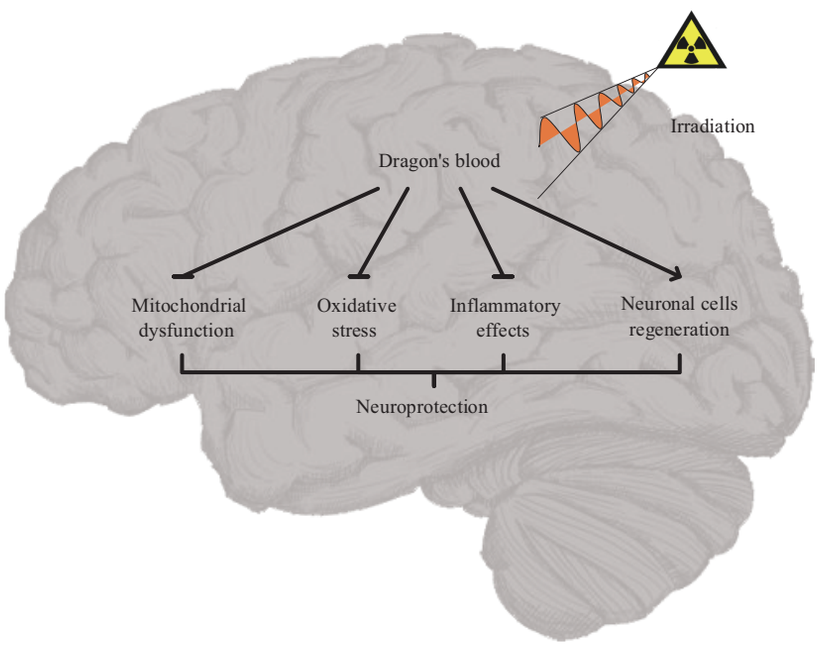

Fig. 1 Protective effects of Dragon's Blood in irradiation−induced neuroinflammation

References

[1] Cucinotta F, Ottolenghi A. Review of Space Radiation Health Risks for Human Exploration on the Moon, Mars and Neos; proceedings of the 42nd COSPAR Scientific Assembly, F, 2018 [C]

[2] Jandial R, Hoshide R, Waters J D, *et al*. Space-brain: The negative effects of space exposure on the central nervous system. Surgical neurology international, 2018, **9**: 9

[3] Kokhan V S, Matveeva M I, Mukhametov A, *et al*. Risk of defeats in the central nervous system during deep space missions. Neuroscience & Biobehavioral Reviews, 2016, **71**: 621-632

[4] Unthank J L, Ortiz M, Trivedi H, *et al*. Cardiac and renal delayed effects of acute radiation exposure: organ differences in vasculopathy, inflammation, senescence and oxidative balance. Radiat Res, 2019, **191**(5): 383-397

[5] Cucinotta F A, Alp M, Sulzman F M, *et al*. Space radiation risks to the central nervous system. Life Sci Space Res, 2014, **2**: 54-69

[6] Lu A P, Jia H W, Xiao C, *et al*. Theory of traditional Chinese medicine and therapeutic method of diseases. World J Gastroenterol, 2004, **10**(13): 1854-1856

[7] Lin C H, Yang A W H, Phillips D, *et al*. An Investigation on Integrating Eastern and Western Medicine with Informatics [M]. Theories to Inform Superior Health Informatics Research and Practice. Springer. 2018: 427-442

[8] Yi T, Chen H B, Zhao Z Z, *et al*. Comparison of the chemical profiles and anti-platelet aggregation effects of two "Dragon's Blood" drugs used in traditional Chinese medicine. Journal of ethnopharmacology, 2011, **133**(2): 796-802

[9] Xin N, Li Y J, Li X, *et al*. Dragon's blood may have radioprotective effects in radiation-induced rat brain injury. Radiat Res, 2012, **178**(1): 75-85

[10] Zhang X R, He Y, Jiang X R, *et al*. Nature brings new avenues to the therapy of central nervous system diseases—An overview of

possible reatments derived from natural products. Sci China Life Sci, 2019, **62**(8): 1-36

[11] Gupta D, Bleakley B, Gupta R K. Dragon's blood: botany, chemistry and therapeutic uses. Journal of ethnopharmacology, 2008, **115**(3): 361-380

[12] Lefaki M, Papaevgeniou N, Chondrogianni N. Redox regulation of proteasome function. Redox Biol, 2017, **13**: 452-458

[13] Irmak M K, Fadillioglu E, Gulec M, *et al*. Effects of electromagnetic radiation from a cellular telephone on the oxidant and antioxidant levels in rabbits. Cell Biochem Funct, 2002, **20**(4): 279-283

[14] Ismail A F, El-Sonbaty S M. Fermentation enhances Ginkgo biloba protective role on gamma-irradiation induced neuroinflammatory gene expression and stress hormones in rat brain. J Photochem Photobiol B, 2016, **158**:154-163

[15] Ran Y, Wang R, Gao Q, *et al*. Dragon's blood and its extracts attenuate radiation-induced oxidative stress in mice. Journal of radiation research, 2014, **55**(4): 699-706

[16] Ran Y, Wang R, Lin F, *et al*. Radioprotective effects of Dragon's blood and its extract against gamma irradiation in mouse bone marrow cells. Phys Med, 2014, **30**(4): 427-431

[17] Xin N, Yang F-J, Li Y, *et al*. Dragon's blood dropping pills have protective effects on focal cerebral ischemia rats model. Phytomedicine, 2013, **21**(1): 68-74

[18] Kim G J, Fiskum G M, Morgan W F. A role for mitochondrial dysfunction in perpetuating radiation-induced genomic instability. Cancer Res, 2006, **66**(21): 10377-10383

[19] Yoshida T, Goto S, Kawakatsu M, *et al*. Mitochondrial dysfunction, a probable cause of persistent oxidative stress after exposure to ionizing radiation. Free Radic Res, 2012, **46**(2): 147-153

[20] Wang Q, Cai H, Hu Z, *et al*. Loureirin B promotes Axon regeneration by inhibiting endoplasmic reticulum stress: induced mitochondrial dysfunction and regulating the Akt/GSK-3β pathway after spinal cord injury. Journal of Neurotrauma, 2019, **36**(12): 1949-1964

[21] Greene-Schloesser D, Robbins M E, Peiffer A M, *et al*. Radiation-induced brain injury: A review. Front Oncol, 2012, **2**:73

[22] Moravan M J, Olschowka J A, Williams J P, *et al*. Cranial irradiation leads to acute and persistent neuroinflammation with delayed increases in T-cell infiltration and CD11c expression in C57BL/6 mouse brain. Radiat Res, 2011, **176**(4): 459-473

[23] Choy C S, Hu C M, Chiu W T, *et al*. Suppression of lipopolysaccharide-induced of inducible nitric oxide synthase and cyclooxygenase-2 by Sanguis Draconis, a dragon's blood resin, in RAW 264.7 cells. Journal of Ethnopharmacology, 2008, **115**(3): 455-462

[24] Son Y, Yang M, Kang S, *et al*. Cranial irradiation regulates CREB-BDNF signaling and variant BDNF transcript levels in the mouse hippocampus. Neurobiol Learn Mem, 2015, **121**:12-19

[25] Kim D H, Zhao X. BDNF protects neurons following injury by modulation of caspase activity. Neurocrit Care, 2005, **3**(1): 71-76

[26] Patrick B. IκB-NF-κB structures: at the interface of inflammation control. Cell, 1998, **95**(12): 729-731

[27] Li Y S, Wang J X, Jia M M, *et al*. Dragon's blood inhibits chronic inflammatory and neuropathic pain responses by blocking the synthesis and release of substance P in rats. J Pharmacol Sci, 2012, **118**(1): 43-54

[28] Sun F, Deng Y, Han X, *et al*. A secret that underlies Parkinson's disease: The damaging cycle. Neurochemistry international, 2019, 104484

[29] Sang X S, Chen Y H, Chen W, *et al*. Celastrol specifically inhibits the activation of NLRP3 inflammasome. Sci China Life Sci, 2017, **60**(7): 1-3

龙血竭对辐射诱导的神经炎症的保护作用*

张　鹏[1)**]　王海龙[1,2)**]　马　宏[1)]　沈乾伟[1)]　孙飞一[1)]
陈　钰[1)]　蓝　钰[1)]　戴荣继[1)]　邓玉林[1)***]

([1)] 北京理工大学生命学院，北京 100081；
[2)] 天津市肿瘤医院肿瘤细胞生物学实验室，天津 300060）

摘要　深空探测的不断发展，潜在的战争威胁以及核事故的发生增加了人类辐射暴露的风险. 神经炎症是人体在辐射暴露后重要生理反应之一. 神经炎症的发生与神经退行性疾病如阿尔茨海默病（Alzheimer's disease，AD）和帕金森病（Parkinson's disease，PD）密切相关. 开发具有抗氧化和抗炎作用的中药对辐射引起的中枢神经系统损伤有积极意义. 中药龙血竭对辐射诱导的神经炎症具有良好的治疗作用. 本文总结了龙血竭在降低氧化应激水平，相关炎症因子表达和线粒体损伤中的作用. 同时提出，内源性神经毒素可能加重辐射诱导的神经炎症的进程，而龙血竭可缓解这种神经炎症.

关键词　龙血竭，辐射，活性氧，神经炎症，线粒体，内源性神经毒素
中图分类号　R741.02，R730.55　　**DOI**：10.16476/j.pibb.2020.0246

* 载人航天工程航天医学实验领域项目（HYZHXM02003）资助.
** 并列第一作者.
*** 通讯联系人.
Tel：010-68915996，E-mail：deng@bit.edu.cn
收稿日期：2020-07-11，接受日期：2020-07-21