RESEARCH HIGHLIGHT

miR-21, a potential therapeutic target of alleviating blood-brain barrier damage after traumatic brain injury

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Received: February 25, 2015
Published online: March 11, 2015

Traumatic brain injury (TBI) is a disease with high morbidity and leads to high rate of disability and mortality. The blood-brain barrier (BBB) damage is an important pathological change in brain after TBI, which impacts the development and prognosis of the disease. In our previous research, we have reported that miR-21-5p in neurons can exert an anti-apoptosis effect, and promote the restoration of injured BBB after TBI. Up-regulation of miR-21-5p level in brain can amplify the above functions of miR-21-5p, thus alleviate BBB damage and improve the neurological prognosis of TBI. miR-21-3p, which is generated from pre-miR-21 at the same time with miR-21-5p, has been reported to play significant roles in regulating cellular apoptosis, immuno-inflammatory responses and the expression of angiogenic factors in various diseases. We made a summary of previous research on miR-21-3p and inferred that miR-21-3p could exert an important impact on BBB damage after TBI. Taken together, miR-21 is a vital miRNA that impacts the restoration of injured BBB, thus it could be a potential therapeutic target for interventions in TBI.

Keywords: traumatic brain injury; miR-21; blood-brain barrier

To cite this article: Xintong Ge, *et al*. miR-21, a potential therapeutic target of alleviating blood-brain barrier damage after traumatic brain injury. RNA Dis 2015; 2: e642. doi: 10.14800/rd.642.

Traumatic brain injury (TBI) is the leading cause of long-term disability in children and young adults. About 10 million people are affected by a new TBI event every year around the world [1]. With the development of urbanization, the increase of traffic accidents and regional wars, the incidence of TBI is increasing rapidly in recent years [2]. Scientists predict that TBI will be the third largest cause of worldwide disease by 2020 [3].

Elucidating the pathological changes involved in TBI and improving the clinical treatment level for TBI has long been important issues for neuroscientists and neurosurgeons. The primary insult leads to not only primary brain damage (including cerebral contusion and laceration, extradural hematoma, subdural hematoma, intracranial hemorrhage, subarachnoid hemorrhage, etc.), but also a series of pathological events like immuno-inflammatory responses and cellular apoptosis/necrosis in brain [4]. These pathological events interact with each other and develop concurrently. They result in secondary brain damage, which mainly includes nerve cells apoptosis, impaired local blood supply and BBB damage [5]. The BBB damage is an important pathological change in brain that it is a significant cause of

serious complications of TBI, like brain edema, intracranial infection and subsequent neurological dysfunction [6]. As the primary insult cannot be therapeutically influenced, promoting the restoration of injured BBB is crucial for treating and improving the prognosis of TBI.

The clinical therapeutic methods for TBI mainly includes oxygen inhalation therapy, decompressive craniectomy, evacuation of intracranial hematoma, dehydrant and neurotrophic drug treatment. As these methods are not aimed at directly alleviating BBB damage, their therapeutic effects are limited in controlling secondary brain damage. The high-dose glucocorticoid pulse therapy was supposed to be effective in alleviating BBB damage after TBI. However, a clinical trail (MRC CRSAH) involved 10008 patients conducted in more than 40 countries demonstrated that corticosteroids does not have any therapeutic effect on TBI patients in spite of the time of administration post-injury [7]. Our previous research also proved that high-dose methylprednisolone can aggravate cellular apoptosis and BBB damage in hypothalamus, which can exacerbate the acute critical illness-related corticosteroid insufficiency associated with TBI [8-9]. Molecular therapy for TBI has become the research hotspot in recent years. The function of erythropoietin, progesterone and other molecules in alleviating BBB damage have been widely confirmed in animal models. However, these therapeutic methods are not effective in TBI patients according to the reports of clinical trials [10, 11]. Therefore, exploring new therapeutic strategies for alleviating BBB damage is crucial for the clinical treatment of TBI.

miR-21 is a non-protein coding RNA molecule that negatively modulate the expression of its target proteins. It has been proved to be an oncogene and implicated in the etiology of a variety of diseases, like cancer and cardiovascular diseases. However, little attention has been paid to their roles in acute diseases, such as TBI. Mature miR-21 includes miR-21-5p and miR-21-3p. They are both generated from pre-miR-21 at the same time through several steps of enzyme digestion [12]. As to the previous research on miR-21-5p, Redell [13] and our group [14] have detected the miRNAs change in brain of TBI rats using miRNAs profiling and qRT-PCR, indicating that the expression level of miR-21-5p in the hippocampus and injured cerebral cortex were both increased from 6 h to 72 h post-injury. PDCD4 and Tiam1 were predicted to be the target genes of miR-21-5p after TBI [14]. These findings suggest that miR-21-5p is a potential biomarker and therapeutic target in TBI. In addition, Sandhir [16] found that miR-21-5p showed no increased expression in response to injury in aging mice following TBI, and the expression of its target genes, including PTEN and PDCD4, were up-regulated. As the activation of PTEN and PDCD4 can promote cellular apoptosis, the diminished miR-21-5p injury response may contribute to the poor prognosis of aged mice compared to the adult. Consequently, strategies aimed at up-regulation of miR-21-5p and/or down regulation of its targets might be useful in improving outcomes following TBI.

Since 2009, we has performed a series of research on the function and mechanism of miR-21 in TBI. We have proved that neurons-expressed miR-21-5p exerted an anti-apoptosis effect in brain after TBI, thereby promoting the restoration of injured BBB and improving the neurological prognosis of TBI [15, 17-19]. Specifically, we employed the fluid percussion injury rat model, and detected the expression level of miR-21-5p in the traumatic focci using qRT-PCR. We found that the miR-21-5p level was increased from 6 h to 72 h post-injury, and it can be further up-regulated or down-regulated by intracerebroventricularly injection of miR-21-5p agomir or antagomir [18]. We then measured the expression of miR-21-5p in neurons, astrocytes, microglias and brain microvascular endothelial cells (BMVECs) respectively using combined miRNA in-situ hybridization and immunofluorescence. We found that compared to astrocytes and microglias, neurons and BMVECs expressed more miR-21-5p, and their miR-21-5p levels are more sensitive in response to injury [18]. We evaluated the influence of miR-21-5p on the neurological outcome of TBI rats using modified neurological severity score and Morris Water Maze. We found that the increased miR-21-5p level in brain post-injury was beneficial for improving the neurological prognosis of TBI [18]. Then, we observed the impact of miR-21-5p on cellular apoptosis in rats and in-vitro cultured neurons to investigate the mechanism of its protective effect in TBI. We confirmed that miR-21 inhibited apoptosis in neurons through inhibiting PTEN expression at the post-transcriptional level and activating Akt signaling [18-19]. In addition, we evaluated the impact of miR-21-5p on BBB damage by measuring brain water content, the amount of Evan's Blue extravasation and the expression level of tight junction proteins in brain. We demonstrated that miR-21-5p can alleviate BBB damage by activating Ang-1/Tie-2 axis. And this effect is also associated with its inhibition on cellular apoptosis in brain [17-18]. Taken together, up-regulation of miR-21-5p level in brain can inhibit the cellular apoptosis and alleviate BBB damage, thus improve the neurological prognosis of TBI.

Recently, the biological function of miRNA* in various diseases began to drawn increasing attention from scientists. As to miR-21-3p (miR-21*), it has been confirmed that its expression is significantly up-regulated in various tumor cells, including non-small cell lung cancer [20], colorectal cancer [21] and several human carcinomas [22]. Besides,

miR-21-3p can activate the Akt signaling that regulates cellular apoptosis in breast tumors [23]. The research on human aortic endothelial cells also found that miR-21-3p is an up-stream regulator of NF-kB signaling that can regulate the immuno-inflammatory responses [24]. In the research on cardiac hypertrophy, miR-21-3p has been proved to have the function of impacting the hypertrophic progress of cradiomyocytes induced by Ang-2 infusion, suggesting that miR-21-3p could regulate the expression of angiogenic factors, such as Ang-1 and Ang-2 [25-26]. Taken together, miR-21-3p may possibly have the function of regulating cellular apoptosis, immuno-inflammatory responses and the expression of angiogenic factors in brain. From this, we inferred that miR-21-3p could exert an important impact on BBB damage after TBI. And we have planned to study the function and mechanism of miR-21-3p in regulating the restoration of BBB damage in the future.

In conclusion, miR-21 is a vital miRNA that impacts the restoration of injured BBB, thus it could be a potential therapeutic target for interventions in TBI. The outcome of our work has further elucidated the mechanism of BBB damage and restoration, and also provided insights into developing a novel therapeutic strategy for TBI.

Conflict of interest

The authors declare no conflict of interest.

Acknowledgements

This work was supported by grants from the National Natural Science Foundation of China (grant no. 81330029, 81471252 and 81271361), Tianjin Municipal Science and Technology Commission natural fund project (grant no. 13JCYBJC23700) and National Clinical Key Subject Construction Project of NHFPC Fund. We also express special acknowledgement to the senior professor Chunsheng Kang who contributes greatly on this work.

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