Noncoding RNA as Diagnostic and Prognostic Biomarkers in Cerebrovascular Disease

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Noncoding RNAs (ncRNAs), such as microRNAs, long noncoding RNAs, and circular RNAs, play an important role in the pathophysiology of cerebrovascular diseases (CVDs). They are effectively detectable in body fluids, potentially suggesting new biomarkers for the early detection and prognosis of CVDs. In this review, the physiological functions of circulating ncRNAs and their potential role as diagnostic and prognostic markers in patients with cerebrovascular diseases are discussed, especially in acute ischemic stroke, subarachnoid hemorrhage, and moyamoya disease.

1. Introduction

Cerebrovascular disease (CVD) is one of the leading causes of disability and mortality worldwide. Recent epidemiological studies have shown that CVD was one of the top ten leading causes of total years of life lost worldwide, especially in China that is of great severity [1, 2]. They mainly include ischemic and hemorrhagic events, but also rare diseases such as moyamoya disease.

The human genome is characterized by a number of noncoding RNAs (ncRNAs) of unknown function. The ncRNAs are involved in many cellular processes and include microRNAs (miRNAs), long noncoding RNAs (lncRNAs), circular RNAs, and transfer RNA-derived small RNAs (tsRNAs). In addition to their intracellular activity, ncRNAs are released within extracellular vesicles in the blood, which makes them potential biomarkers in different pathological conditions [3]. Several studies have reported that circulating ncRNAs could be measured both in tissues and in biological fluids, supporting their potential use as diagnostic and prognostic markers [4–8].

In this review, we discuss the physiological functions of circulating ncRNAs and their potential role as diagnostic markers, prognostic markers, and therapeutic targets in patients with cerebrovascular diseases, including acute ischemic stroke (AIS), subarachnoid hemorrhage (SAH), and moyamoya disease (MMD). Current challenges and future perspectives are also discussed.

2. Biogenesis and Transport of ncRNAs

A large number of studies have demonstrated that CVDs may affect the expression level of ncRNAs in body fluids. A multitude of ncRNAs have been described in body fluids, such as serum, plasma, urine, and breast milk [9]. Different techniques have been used, including RNA sequencing, microarray screening, and real-time quantitative polymerase chain reaction (RT-qPCR) [10].

Apparently, the ncRNAs existing in body fluids mostly come from specific cells, tissues, or organs, which are quite relevant to the disease conditions, whereas there are only a few studies that provided potential mechanisms involving ncRNAs in some disease conditions. So far, five transport mechanisms of ncRNA have been described (Figure 1): (1) exosomes, (2) microparticles, (3) apoptotic bodies, (4) ribonucleoproteins, including argonaute-2 (AGO2), nucleophosmin-
1 (NPM1), and high-density lipoproteins (HDLs), and (5) direct cellular connections, such as gap junctions, synapses, and intercellular bridges [10–16]. After secretion to the extracellular space, ncRNAs target specific cells and organs and exercise certain functions. Extracellular vesicles, like exosomes, microparticles, and apoptotic bodies, can transfer cargos from parental cells to recipient cells, achieving cell-to-cell communication, whereas the exact process how these extracellular vesicles are able to recognize the target cells remains unknown [10]. Ribonucleoproteins are other important carriers of ncRNAs. More than 90% of circulating miRNAs transfer through this pathway [10, 17]. The ncRNAs are quite stable and protected from RNases in body fluids, indicating that they are promising biomarkers in assessing the pathophysiological changes in the body. However, further studies are required to clarify the relationship between circulating ncRNAs and their carriers, how they interact with target cells, and what functions they exert [18].

3. ncRNA in Acute Ischemic Stroke

Effective management of patients with acute cerebrovascular disease relies on precise diagnosis and timely treatment, especially those with acute ischemic stroke (AIS). AIS accounts for around 80% of strokes and only has a narrow therapeutic window [19]. However, the accurate diagnosis of AIS can be disturbed by stroke mimics and other types of strokes. A computed tomography (CT) scan is usually able to detect a stroke from a blood clot or bleeding within the brain, but around half of patients are false negatives [20, 21]. MR diffusion-weighted imaging (DWI) is highly sensitive in detecting and localizing acute ischemic brain lesions but is limited by the length of the procedure, the lack of availability in remote areas, expensive costs, and prehospital settings [22]. Blood-based biomarkers with high sensitivity and specificity are therefore attractive, but those available to date have poor diagnostic accuracy [23].

Several questions in AIS diagnosis and treatment that remain to be solved can be briefly concluded as follows: the first is how to diagnose the AIS with high effectiveness and precision, which means that physicians should not only identify AIS immediately but also exclude other differential diagnoses like ICH, SAH, and stroke mimic confidently and further ascertain the stroke etiology (TOAST type) to instruct therapeutic strategies. In this setting, highly sensitive and specific biomarkers are needed in clinical practice. The second one is how to evaluate the short-term and long-term outcomes after stroke, which can be beneficial for planning subsequent therapeutic strategies and decide whether an aggressive treatment is helpful to promote post-stroke recovery. To answer these questions, various investigations that focus on the ncRNA-based biomarkers are summarized as follows.

3.1. miRNAs as Diagnostic Biomarkers in AIS. Circulating miRNAs have been reported as potential diagnostic and prognostic biomarkers in several studies (Table 1). Using RNA sequencing data, miR-125a-5p, miR-125b-5p, and
Table 1: The studies about ncRNA as diagnostic and prognostic biomarkers in AIS patients.

| Reference        | Sample size | ncRNA          | Platform          | Sample         | Findings                                                                                                                                 |
|------------------|-------------|----------------|-------------------|----------------|------------------------------------------------------------------------------------------------------------------------------------------|
| Long et al. [27] | 50 HC and 197 AIS | miR-30a, miR-126, let-7b | RT-qPCR          | Plasma         | miR-30a and miR-126 were significantly downregulated in AIS patients, while let-7b was downregulated in large-artery atherosclerosis but upregulated in other kinds of AIS patients |
| Leung et al. [28] | 79 AIS and 19 ICH | miR-124-3p, miR-16 | RT-qPCR          | Plasma         | miR-124-3p was significantly upregulated in ICH patients, while miR-16 was significantly upregulated in AIS patients |
| Wu et al. [25]  | 120 HC and 106 AIS | miR-15, miR-16, miR-17-5p | RT-qPCR          | Serum          | These 3 miRNAs were significantly upregulated in AIS patients                                                                                                                                   |
| Wang et al. [31] | 58 HC and 58 AIS | miR-29b         | RT-qPCR          | Peripheral leukocyte | miR-29b was downregulated in AIS patients and was negatively associated with the NIHSS score and infarct volume |
| Rainer et al. [35] | 51 AIS | miR-124-3p, miR-16 | RT-qPCR          | Plasma         | Higher miR-124-3p was associated with mRS > 2 and mortality within 90 days; miR-16 was the opposite |
| Huang et al. [36] | 38 HC and 76 AIS | miR-132        | RT-qPCR          | Serum          | miR-132 was higher in poststroke cognitive impairment                                                                                     |
| Dykstra-Aiello et al. [39] | 133 HC and 133 AIS | —              | Microarray       | Whole blood    | IncRNA expression profiling changes after stroke and can change over time                                                               |
| Sørensen et al. [37] | 21 HC and 21 AIS | miR-9-5p, miR-9-3p, miR-124-3p, miR-128-3p | RNA-seq and RT-qPCR | CSF            | miR-9-5p, miR-9-3p, miR-124-3p, and miR-128-3p were higher in bigger infarct size patients                                                  |
| Scherrer et al. [32] | 329 AIS | miR-150-5p      | RT-qPCR          | Plasma         | Lower miR-150-5p was significantly associated with mortality within 90 days                                                             |
| Jin and Xing [33] | Discovery: 10 HC and 10 AIS | miR-126, miR-378, miR-101, miR-222, miR-218, miR-206 | RT-qPCR          | Plasma         | miR-126, miR-378, miR-101, miR-222, miR-218, and miR-206 were associated with the NIHSS score                                             |
| Chen et al. [34] | 33 HC and 50 AIS | miR-223         | RT-qPCR          | Exosome         | Uregulated miRNA was associated with the high NIHSS score and mRS > 2 within 90 days                                                        |
| Tiedt et al. [24] | Discovery: 20 HC and 20 AIS | miR-125a-5p, miR-125b-5p, miR-143-5p | RNA-seq in the discovery cohort; RT-qPCR in the validation and republication cohort | Plasma         | These 3 miRNAs’ upregulation can identify the AIS from HC and TIA patients                                                              |
| Wang et al. [26] | 39 HC and 78 AIS | miR-221-3p, miR-382-5p | RT-qPCR          | Serum         | These 2 miRNAs were significantly downregulated in AIS patients                                                                          |
| Zhu et al. [40]  | 189 HC and 189 AIS | IncRNA MIAT    | RT-qPCR          | Peripheral leukocyte | Uregulated MIAT was associated with NIHSS scores, mRS, high-sensitivity C-reactive protein, and infarct volume                        |
| Gui et al. [30]  | Discovery: 13 HC and 87 AIS | miR-125b, miR125a, let-7b, let-7e, miR-7-2-3p, miR-1908 | Microarray in the discovery cohort; RT-qPCR in the validation cohort | Serum         | miRNAs were associated with the TOAST subtype                                                                                           |
miR-143-3p were constructed to discriminate between AIS and transient ischemic attack (TIA) patients and healthy controls. Longitudinal analysis showed a significant increase in miR-125a-5p, miR-125b-5p, and miR-143-3p during the first 24 hours after AIS, with miR-125b-5p and miR-143-3p returning to normal levels within 48 hours after AIS, which reveals a good discrimination in the acute phase. A random forest classification model presented good performance in differentiating AIS and healthy controls [24]. Another study showed that serum miR-15a, miR-16, and miR-17-5p significantly increased in patients with AIS compared to healthy controls [25]. Similarly, Wang et al. reported that serum miRNA-221-3p and miRNA-382-5p were downregulated in patients with AIS [26]. The downregulation of plasma miR-30a, miR-126, and let-7b was also described in patients with AIS [27].

The miRNAs are also thought to identify stroke subtypes (Table 1). As reported by Leung et al., miR-124-3p and miR-16 are potential diagnostic biomarkers between intracerebral hemorrhage (ICH) and AIS. Plasma concentration of miR-124-3p was significantly higher, whereas the concentration of miR-16 was significantly lower in patients with ICH than in AIS patients within 24 hours after the stroke onset [28]. These findings indicated that plasma miRNAs have the potential to distinguish ICH from AIS. Kalani et al. comprehensively profiled miRNAs across acute stroke subtypes through next-generation sequencing. The sequencing data were put into LASSO regression analysis to classify AIS, ICH, and subarachnoid hemorrhage (SAH) [29]. In discriminating different ischemic stroke subtypes, miRNA can also act as an effective biomarker. A total of 87 patients with AIS and 13 healthy subjects were recruited. The ROC analysis demonstrated that miR-125b, miR-125a, let-7b, and let-7e discriminate between acute stroke due to cardiac embolism and other subtypes of stroke. Besides, miR-7-2-3p and miR-1908 showed significant AUC in both large-artery atherosclerosis and lacunar infarct patients [30].

### 3.2. miRNAs as Prognostic Biomarkers in AIS

The predictive value of miRNAs in patients with AIS is shown in Table 1. Wang et al. reported that miR-29b was significantly downregulated in patients with AIS and negatively associated with National Institute of Health Stroke Scale (NIHSS) scores and stroke volume. Of note, the overexpression of miR-29b reduced the infarct volume and brain edema and infarct volume of the brain in mice [31]. In a prospective cohort study, miRNA-150-5p was negatively associated with mortality in patients with AIS within 90 days after the stroke onset [32]. Some miRNAs related to angiogenesis, including miR-126, miR-378, and miR-101, were negatively correlated with NIHSS scores, while miR-222, miR-218, and miR-206 were positively associated with NIHSS scores [33]. The exosomal miR-223 was positively associated with AIS occurrence, stroke severity, and short-term outcomes [34]. In another study conducted by Rainer et al., plasma miR-124-3p was elevated in patients with AIS who died within 3 months after hospital admission, while miR-16 was better associated with survival [35]. A high level of serum miR-132 correlated with post-stroke cognitive impairment [36], while several brain-enriched miRNAs (miR-9-5p, miR-9-3p, miR-124-3p, and miR-128-3p) were elevated in patients with infarcts larger than 2 cubic meters [37]. Overall, several miRNAs are potential biomarkers associated with the prognosis of AIS.

### 3.3. Other Biomarkers in AIS

Other ncRNAs also play an important role in diagnostic and prognostic assessment of AIS (Table 1). Recently, Zuo et al. suggested that increased plasma levels of circFUNDC1, circPDSSB, and circCDC14A may be useful to diagnose AIS. In addition, an opposite trend was observed in patients with good modified Rankin Scale (mRS) scores within 7 days, suggesting their prognostic values [38]. Like other ncRNAs, lncRNAs may change in patients with AIS. In a case-control study, a large array of differentially expressed IncRNAs were detected in blood. Specifically, IncRNA NR-002332 and IncRNA A131606 were upregulated, while IncRNA C10 and IncRNA F57-2 were downregulated [39]. At the same time, IncRNAs in the blood were associated with clinical outcomes: the IncRNA MIAT was significantly upregulated and correlated with the NIHSS score, mRS score, serum C-reactive protein, and infarct volume [40].

### 3.4. ncRNA as a Therapeutic Target in AIS

The secondary brain injuries are common problems after AIS, such as brain...
edema, ischemic reperfusion injury, and hemorrhagic transformation. Recent studies have demonstrated that ncRNAs make great contribution to exacerbate or attenuate these injury processes through affecting neuroinflammation, neural apoptosis, oxidative stress, microglia activation, and excitotoxicity. Besides, neuroprotective ncRNA or their mimics can serve as promising drugs for their ability to penetrate the blood-brain barrier (BBB). Therefore, ncRNA may play a crucial role in improving AIS outcomes when serving as the therapeutic target. For example, ncRNA can influence the AIS process through regulating the oxidative stress process [41]. The upregulation of miR-424 pre- and poststroke in middle cerebral artery occlusion (MCAO) mice can decrease the cerebral infarction volume and brain edema by inhibiting oxidative stress, cellular apoptosis, and microglia activation [42], while miR-106b-5p antagonist could also decrease the infarction volume and neurological deficit in rats by regulating oxidative stress after AIS [43]. Other regulation of ncRNAs can also attenuate the oxidative stress following ischemic stroke. For example, the downregulation of miR-93 and miR-182 or the upregulation of miR-23a-3p, and miR-99a protected the AIS brain [44–47]. miR-93 antagonist alleviates ischemic injury through the Nrf2/HO-1 antioxidant pathway [47]. miR-182 promoted nitric oxide (NO) and 3-nitrotyrosine (3-NT) production and caspase-3 expression, while reducing superoxide dismutase (SOD) and manganese SOD (MnSOD) activities [46]. On the contrary, miR-23a-3p attenuated oxidative stress injury by reducing the production of NO and 3-NT and increasing the production of SOD and MnSOD [44]. All these researches indicated that miRNA might be a promising therapeutic target by attenuating the oxidative stress process.

Other miRNAs, such as miR-223 [48], miR-29b [49], miR-29c [50], miR-17-92 [51], miR-124 [52], miR-210 [53], miR-139-5p [54], miR-let-7c-5p [55], miR-107 [56], miR-207 [57], miR-335 [58], miR-22 [59], miR-9 [60], miR-378 [61], miR-122 [62], miR-210 [63], miR-455 [64], and miR-363 [65], can also reduce infarction volume and improve outcomes in animal models through various mechanisms. For example, miR-223 lowered the levels of glutamate receptors to reduce excitotoxicity and has a therapeutic role after stroke [48]. miR-139-5p agomir reduced neural apoptosis by inhibiting human growth transformation-dependent protein (HGTDP-P), providing a new therapeutic insight [54].

On the contrary, the overexpression of miR-145 [66], miR-320a [67], miR-497 [68], miR-let-7f [69], miR-181a [70], miR-181b [71], miR-103-1 [72], miR-30a [73], miR-124 [74], miR-134 [75], miR-200c [76], miR-155 [77], miR-24 [78], miR-182 [46], miR-493 [79], miR-383 [80], miR-106b-5p [43], miR-15a/16-1 [81], miR-30d-5p [82], miR-337 [83], and miR-337-3p [84] exacerbates the infarction volumes, edema, and neuroinflammation. For instance, inhibition of miR-377 decreased cerebral infarct volume and suppressed cerebral inflammation but promoted angiogenesis in MCAO rats [83]. Reducing poststroke miR-200c was also a potential target to mitigate infarction volume and neurological deficit by inducing reelin expression in mice [76].

4. ncRNAs in Aneurysmal Subarachnoid Hemorrhage

Subarachnoid hemorrhage (SAH) is a medical emergency, accounting for 2–7% of all stroke cases, mostly due to aneurysm rupture in over 80% of cases [85]. The diagnosis of SAH mainly relies on CT scan. If the CT scan is not definitive, the next recommended diagnostic tool is the lumbar puncture [86]. Besides, SAH is also a life-threatening disease with a case fatality of 25–35%, most of which results from the subsequent cerebral vasospasm (CVS) and delayed cerebral infarction (DCI) who survive at the first bleeding event [85–89]. Therefore, the development of a predictive biomarker would be helpful to prevent the development of CVS and understand the precise mechanism behind CVS. The emerging ncRNAs suggest future developments in the diagnostic and prognostic assessment of SAH.

4.1. ncRNAs as Diagnostic Biomarkers in SAH. Several studies have demonstrated that the ncRNA signature in SAH was quite different from that without SAH (Table 2). A microarray analysis and RT-qPCR were utilized to confirm the association among health controls, SAH with DCI, and SAH without DCI. This single study demonstrated that serum miR-132 and miR-324 were upregulated in SAH, compared with healthy controls, while the differences between DCI and non-DCI were not statistically significant. A possible reason was that the sample size was insufficient [90]. Using a similar methodology, Lai et al. reported that miR-502-5p, miR-1297, and miR-4320 were overexpressed in patients with SAH. Seven days after diagnosis, serum miR-502-5p and miR-1297 were significantly higher in patients with SAH. Additionally, at the 7th day after SAH, serum miR-502-5p and miR-1297 levels were significantly higher in patients with increased higher World Federation of Neurological Surgeons (WFNS) and mRS at the ninth month after stroke, which can represent the worse progression of SAH [91]. Another research also determined that serum miR-1297 was directly associated with a higher WFNS grade, Hunt-Hess grade, higher Fisher score, and poor one-year outcome [92]. In cerebrospinal fluid, miR-92a and let-7b decreased, whereas miR-491 increased over time in patients with SAH [93].

4.2. ncRNAs as Prognostic Biomarkers in SAH. With the help of the differential expression profile of ncRNA in SAH patients’ body fluids, ncRNAs are simultaneously competent to distinguish or predict some severe complications after SAH (Table 2). Styli et al. reported that miR-27a-3p, miR-516a-5p, miR-566, and miR-1197 were expressed in cerebrospinal fluid (CSF) differently between patients with cerebral vasospasm (CVS) and those without [94]. In another study, Lu et al. described that 4 circulating miRNAs (miR-4532, miR-4463, miR-1290, and miR-4793) differentiated patients with SAH with delayed cerebral infarction (DCI) from those without DCI by using a machine learning method [95]. A further prospective case-control study demonstrated that an array of miRNA profiles were overexpressed in the CSF of patients with SAH compared with healthy controls. Of interest, the angiographic CVS after SAH was associated
with an increase in miR-132-3p, -19b-3p, -210-3p, -221-3p, and -484 [96], miR-15a and Kruppel-like factor 5 (KLF5), a potent modulator of miR-15a expression, may also be involved in CVS [97]. Several lncRNAs were also investigated in patients with SAH. According to Pan et al., lncRNAs ZFAS1 and MALAT1 were significantly upregulated, whereas lncRNAs LINC00261 and LINC01619 were downregulated in patients with SAH and CVS compared with those without CVS. Moreover, two lncRNAs (MALAT1 and LINC01619) accurately predicted CVS in around 90% of cases [98].

4.3. ncRNA as a Therapeutic Target in SAH. Several investigations have demonstrated that regulation of ncRNA influences many pathophysiological processes after SAH, including apoptosis, autophagy, neuroinflammation, and brain edema. Therefore, ncRNAs serve as promising therapeutic targets in SAH by regulating these underlying processes. For example, circulting exosomal miR-193b-3p treatment suppressed the expression and activity of HDAC3, reducing inflammation reaction in mice after SAH [99]. Intracerebroventricular injection of miR-103-3p antagonist before SAH reduced BBB permeability and improved neurological function [100]. Additionally, by downregulating iNOS and inhibiting the NF-κB signaling pathway, miR-195-5p attenuated white matter injury and SAH-induced vasospasm [101]. Extracellular vesicle derived from the mesenchymal stem cell could transfer miR-21 to neurons, promoting neuronal survival and improving cognitive function after SAH [102]. Liang et al. showed that, in a rat SAH model, lncRNA MEG3 overexpression increased cell apoptosis [103]. Besides, other studies demonstrated that the regulation of miR-26b [104], miR-706 [105], miR-206 [106], miR-675, and let-7a [107] could also affect the brain injury and outcomes after SAH. These results might provide a deeper understanding of the pathophysiological processes in brain injury after SAH, as well as potential therapeutic targets for the translational researches.

5. ncRNA Biomarkers in Moyamoya Disease

Moyamoya disease (MMD) is a rare, chronic, and progressive disorder of blood vessels in the brain. MMD is characterized by progressive occlusion of the internal carotid artery or its terminal branches, associated with the formation of collateral vessels at the base of the brain [108]. It is associated with vascular cognitive impairment [109, 110] and an increased risk of stroke [111, 112]. Although the diagnosis and treatment of MMD are of high standard, the efficiency still needs improvement. According to the current guidelines from Japan [113], the diagnosis requires an angiogram. The diagnosis is mainly characterized by the morphological abnormalities of cerebral arteries but not etiological or pathogenetic abnormalities [114]. Thus, diagnostic biomarkers that can reflect the disease process are eagerly awaited.

Table 2: The studies about ncRNA as diagnostic and prognostic biomarkers in SAH patients.

| Reference               | Sample size | ncRNA                 | Platform                  | Sample | Findings                                                                 |
|-------------------------|-------------|-----------------------|---------------------------|--------|--------------------------------------------------------------------------|
| Su et al. [90] 2015     | 20 HC and 40 SAH | miR-132, miR-324     | Microarray and RT-qPCR    | Serum  | miR-132 and miR-324 were higher in SAH patients                         |
| Powers et al. [93] 2016 | 8 SAH       | —                     | NanoString array and RT-qPCR | CSF    | miRNA expression pattern changed over time after SAH                    |
| Lai et al. [91] 2017    | 10 HC and 60 SAH | miR-502-5p, miR-1297, miR-4320 | Microarray and RT-qPCR    | Serum  | miR-502 and miR-1297 were associated with WFNS and mRS at 9 months      |
| Styli et al. [94] 2017  | 4 HC and 20 SAH | miR-27a-3p, miR-516a-5p, miR-566, and miR-1197 | NanoString array | CSF    | These miRNAs were differentially expressed between SAH patients with and without CVS |
| Lu et al. [95] 2017     | 20 HC and 40 HC | miR-4532, miR-4463, miR-1290, and miR-4793 | RT-qPCR                  | Plasma | 4-miRNA characterized SAH patients with DCI                            |
| Bache et al. [96] 2017  | 10 HC and 27 SAH | miR-21 and miR-221   | High-throughput RT-qPCR   | CSF    | 2 miRNAs upregulated in SAH with DCI                                    |
| Kikkawa et al. [97] 2017| 3 HC and 8 SAH | miR-15a               | Microarray and RT-qPCR    | Plasma | The dysregulation of miR-15a and KLF4 after SAH may result in CVS       |
| Sheng et al. [92] 2018  | 40 HC and 128 SAH | miR-1297             | RT-qPCR                  | Serum  | miR-1297 was associated with WFNS, Hun-Hess grade, and Fisher score and 1-year mRS |
| Pan et al. [98] 2020    | Discovery: 10 HC and 20 SAH Validation: 65 SAH with and without CVS | IncRNA MALAT1, lncRNA LINC01619 | RT-qPCR                  | CSF    | These two lncRNA signatures can predict the occurrence of CVS             |

Abbreviation: CSF: cerebrospinal fluid; CVS: cerebral vasospasm; DCI: delayed cerebral infarction; HC: health control; mRS: modified Rankin Scale; RT-qPCR: real-time quantitative polymerase chain reaction; SAH: subarachnoid hemorrhage; WFNS: World Federation of Neurological Surgeons.
### 6. Current Challenges and Future Perspectives

Although several ncRNA biomarkers are currently rapidly developing, the use of ncRNA as effective biomarkers in the clinical settings still has to face some unavoidable challenges. Several ncRNAs have been tested as biomarkers in cerebrovascular diseases. They have achieved some success in distinguishing sick people from healthy controls, but sample size in these studies was relatively small and the ability of ncRNAs to discriminate between different disease subtypes or other similar diseases needs further study. At present, the sensitivity, specificity, and reproducibility of ncRNAs are not at their best. Furthermore, the standardization of sampling and testing specimens collected from different body fluids requires more investigation, since different blood centrifugation conditions, sample storage conditions, sequencing platforms, and ncRNA isolation kits can affect the outcomes [120–122]. Moreover, patients from real-life conditions have several comorbidities that may influence the expression of circulating ncRNAs, such as hypertension, diabetes mellitus, and cancer [123, 124]. Even different lifestyles, like smoking or dietary structures, affect the expression level of circulating miR-126, miR-130a, and miR-92a, which may affect the diagnosis of coronary artery disease when using miR-126 as a ncRNA-based biomarker [125, 126]. Additionally, the exact regulatory mechanisms of ncRNA and their physical functions are not clear. The analysis of confounding factors that affect ncRNA expression would be difficult.

Despite the shortcomings mentioned before, ncRNA biomarkers are still promising biomarkers in cerebrovascular diseases. The first ncRNA-based biomarker, IncRNA prostate cancer antigen 3, approved by FDA in 2012, has been routinely utilized in prostate cancer diagnosis, which stimulated the further development of ncRNA biomarkers in other diseases [127, 128]. ncRNAs are abundant and easily detectable in body fluids and are especially appealing as biomarkers because they are not prone to RNase degradation and remain stable in stored samples [129–131].

With the rapid development of artificial intelligence and machine learning (ML), we have never been closer to help physicians making data-driven medical decisions. The concentration of circulating ncRNAs may become essential to diagnose disease and predict outcomes. Several attempts have been made to determine their potential as biomarkers,
using random forest algorithms, support vector machines, and LASSO [4–8, 24]. However, the significance of ncRNAs in cerebrovascular diseases remains poor. Further studies using ML are ongoing and will shed new light on the topic.

This review discusses the important researches about the present situation and the advance in diagnostic and prognostic ncRNA biomarkers of several cerebrovascular diseases. Although the researches of ncRNA in cerebrovascular disease remain at the preclinical stage, such studies gave us clues of understanding cerebrovascular pathophysiology and finally would drive us to a more accurate diagnosis and prognosis for cerebrovascular diseases. Further studies using ML are ongoing and will shed new light on the topic.

**Conflicts of Interest**

The authors declare that there is no conflict of interest regarding the publication of this article.

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**References**

[1] “Global, regional, and national age-sex specific mortality for 264 causes of death, 1980–2016: a systematic analysis for the Global Burden of Disease Study 2016,” Lancet, vol. 390, no. 10100, pp. 1151–1210, 2017.

[2] S. Wu, B. Wu, M. Liu et al., “Stroke in China: advances and challenges in epidemiology, prevention, and management,” Lancet Neurology, vol. 18, no. 4, pp. 394–405, 2019.

[3] H. Valadi, K. Ekström, A. Bossios, M. Sjöstrand, J. J. Lee, and J. O. Lötvall, “Exosome-mediated transfer of mRNAs and microRNAs is a novel mechanism of genetic exchange between cells,” Nature Cell Biology, vol. 9, no. 6, pp. 654–659, 2007.

[4] N. Ludwig, T. Fehlmann, F. Kern et al., “Machine learning to detect Alzheimer’s disease from circulating non-coding RNAs,” Genomics, Proteomics & Bioinformatics, vol. 17, no. 4, pp. 430–440, 2019.

[5] T. Chen, C. Zhang, Y. Liu et al., “A gastric cancer LncRNAs model for MSI and survival prediction based on support vector machine,” BMC Genomics, vol. 20, no. 1, p. 846, 2019.

[6] S. Moustafa, M. Burn, R. Mamillapalli, S. Nematian, V. Flores, and H. S. Taylor, “Accurate diagnosis of endometriosis using serum microRNAs,” American Journal of Obstetrics and Gynecology, vol. 223, no. 4, pp. 557.e1–557.e11, 2020.

[7] M. H. Shellman and Y. G. Shellman, “Human against machine? Machine learning identifies microRNA ratios as biomarkers for melanoma,” The Journal of Investigative Dermatology, vol. 140, no. 1, pp. 18–20, 2020.

[8] L. Yoffe, A. Polsky, A. Gilmot et al., “Early diagnosis of gestational diabetes mellitus using circulating microRNAs,” European Journal of Endocrinology, vol. 181, no. 5, pp. 565–577, 2019.

[9] J. A. Weber, D. H. Baxter, S. Zhang et al., “The microRNA spectrum in 12 body fluids,” Clinical Chemistry, vol. 56, no. 11, pp. 1733–1741, 2010.

[10] J. V. Fritz, A. Heintz-Buschart, A. Ghosal et al., “Sources and functions of extracellular small RNAs in human circulation,” Annual Review of Nutrition, vol. 36, no. 1, pp. 301–336, 2016.

[11] J. D. Arroyo, J. R. Chevillet, E. M. Kroh et al., “Argonaute2 complexes carry a population of circulating microRNAs independent of vesicles in human plasma,” Proceedings of the National Academy of Sciences of the United States of America, vol. 108, no. 12, pp. 5003–5008, 2011.

[12] A. Turchinovich, L. Weiz, A. Langheinz, and B. Burwinkel, “Characterization of extracellular circulating microRNA,” Nucleic Acids Research, vol. 39, no. 16, pp. 7223–7233, 2011.

[13] J. Wagner, M. Riwanto, C. Besler et al., “Characterization of levels and cellular transfer of circulating lipoprotein-bound microRNAs,” Arteriosclerosis, Thrombosis, and Vascular Biology, vol. 33, no. 6, pp. 1392–1400, 2013.

[14] M. Mittelbrunn and F. Sánchez-Madrid, “Intercellular communication: diverse structures for exchange of genetic information,” Nature Reviews. Molecular Cell Biology, vol. 13, no. 5, pp. 328–335, 2012.

[15] P. K. Lim, S. A. Bliss, S. A. Patel et al., “Gap junction-mediated import of microRNA from bone marrow stromal cells can elicit cell cycle quiescence in breast cancer cells,” Cancer Research, vol. 71, no. 5, pp. 1550–1560, 2011.

[16] M. Mittelbrunn, C. Gutiérrez-Vázquez, C. Villarroya-Beltró et al., “Unidirectional transfer of microRNA-loaded exosomes from T cells to antigen-presenting cells,” Nature Communications, vol. 2, no. 1, p. 282, 2011.

[17] K. Wang, S. Zhang, J. Weber, D. Baxter, and D. J. Galas, “Export of microRNAs and microRNA-protective protein by mammalian cells,” Nucleic Acids Research, vol. 38, no. 20, pp. 7248–7259, 2010.

[18] J. Vierreck, C. Bang, A. Foinquinos, and T. Thum, “Regulatory RNAs and paracrine networks in the heart,” Cardiovascular Research, vol. 102, no. 2, pp. 290–301, 2014.

[19] C. C. Beal, “Gender and stroke symptoms,” The Journal of Neuroscience Nursing, vol. 42, no. 2, pp. 80–87, 2010.

[20] J. M. Wardlaw and O. Mielke, “Early signs of brain infarction at CT: observer reliability and outcome after thrombolytic treatment—systematic review,” Radiology, vol. 235, no. 2, pp. 444–453, 2005.

[21] J. Hopyan, A. Ciaramello, D. Dowlatshahi et al., “Certainty of stroke diagnosis: incremental benefit with CT perfusion over noncontrast CT and CT angiography,” Radiology, vol. 255, no. 1, pp. 142–153, 2010.

[22] P. J. Hand, J. M. Wardlaw, C. S. Rivers et al., “MR diffusion-weighted imaging and outcome prediction after ischemic stroke,” Neurology, vol. 66, no. 8, pp. 1159–1163, 2006.

[23] S. Misra, A. Kumar, P. Kumar et al., “Blood-based protein biomarkers for stroke differentiation: a systematic review,” Proteomics. Clinical Applications, vol. 11, no. 9–10, 2017.

[24] S. Tiedt, M. Prestel, R. Malik et al., “RNA-Seq identifies circulating miR-125a-5p, miR-125b-5p, and miR-143-3p as potential biomarkers for acute ischemic stroke,” Circulation Research, vol. 121, no. 8, pp. 970–980, 2017.

[25] J. Wu, K. Du, and X. Lu, “Elevated expressions of serum miR-15a, miR-16, and miR-17-5p are associated with acute ischemic stroke,” International Journal of Clinical and Experimental Medicine, vol. 8, no. 11, pp. 21071–21079, 2015.
[26] Y. Wang, Z. Ma, P. Kan, and B. Zhang, “The diagnostic value of serum miRNA-221-3p, miRNA-382-5p, and miRNA-4271 in ischemic stroke,” Journal of Stroke and Cerebrovascular Diseases, vol. 26, no. 5, pp. 1055–1060, 2017.

[27] G. Long, F. Wang, H. Li et al., “Circulating miR-30a, miR-126 and let-7b as biomarker for ischemic stroke in humans,” BMC Neurology, vol. 13, pp. 1–10, 2013.

[28] L. Y. Leung, C. P. Y. Chan, Y. K. Leung et al., “Comparison of miR-124-3p and miR-16 for early diagnosis of hemorrhagic and ischemic stroke,” Clinica Chimica Acta, vol. 433, pp. 139–144, 2014.

[29] M. Y. S. Kalani, E. Alsop, B. Meechovet et al., “Extracellular microRNAs in blood differentiate between ischemic and haemorrhagic stroke subtypes,” Journal Of Extracellular Vesicles, vol. 9, no. 1, article 1713540, 2020.

[30] Y. Gui, Z. Xu, T. Jin et al., “Using extracellular circulating microRNAs to classify the etiologic subtypes of ischemic stroke,” Translational Stroke Research, vol. 10, no. 4, pp. 352–361, 2019.

[31] Y. Wang, J. Huang, Y. Ma et al., “MicroRNA-29b is a therapeutic target in cerebral ischemia associated with aquaporin 4,” Journal of Cerebral Blood Flow and Metabolism, vol. 35, no. 12, pp. 1977–1984, 2015.

[32] N. Scherrer, F. Fays, B. Mueller et al., “MicroRNA 150-5p improves risk classification for mortality within 90 days after acute ischemic stroke,” Journal Of Stroke, vol. 19, no. 3, pp. 323–332, 2017.

[33] F. Jin and J. Xing, “Circulating pro-angiogenic and anti-angiogenic microRNA expressions in patients with acute ischemic stroke and their association with disease severity,” Neurological Sciences, vol. 38, no. 11, pp. 2015–2023, 2017.

[34] Y. Chen, Y. Song, J. Huang et al., “Increased circulating exosomal miRNA-223 is associated with acute ischemic stroke,” Frontiers in Neurology, vol. 8, article 57, 2017.

[35] T. H. Rainer, L. Y. Leung, C. P. Y. Chan et al., “Plasma miR-124-3p and miR-16 concentrations as prognostic markers in acute stroke,” Clinical Biochemistry, vol. 49, no. 9, pp. 663–668, 2016.

[36] J. Zhao, D. Huang, L. Zhuo, S. Liao, and Z. Jiang, “Serum miR-132 is a risk marker of post-stroke cognitive impairment,” Neurosci Letters, vol. 615, pp. 102–106, 2016.

[37] S. S. Stetens, A. B. Nygaard, A. L. Carlsen, N. H. H. Heegaard, M. Bak, and T. Christensen, “Elevation of brain-enriched miRNAs in cerebrospinal fluid of patients with acute ischemic stroke,” Biomarker Research, vol. 5, no. 1, p. 24, 2017.

[38] L. Zuo, L. Zhang, J. Zu et al., “Circulating circular RNAs as biomarkers for the diagnosis and prediction of outcomes in acute ischemic stroke,” Stroke, vol. 51, no. 1, pp. 319–323, 2020.

[39] C. Dykstra-Aiello, G. C. Jickling, B. P. Ander et al., “Altered expression of long noncoding RNAs in blood after ischemic stroke and proximity to putative stroke risk loci,” Stroke, vol. 47, no. 12, pp. 2896–2903, 2016.

[40] M. Zhu, N. Li, P. Luo et al., “Peripheral blood leukocyte expression of IncRNA MIAT and its diagnostic and prognostic value in ischemic stroke,” Journal of Stroke and Cerebrovascular Diseases, vol. 27, no. 2, pp. 326–337, 2018.

[41] A. Esquela-Kerscher and F. J. Slack, “Oncomirs - microRNAs with a role in cancer,” Nature Reviews. Cancer, vol. 6, no. 4, pp. 259–269, 2006.

[42] P. Liu, H. Zhao, R. Wang et al., “MicroRNA-424 protects against focal cerebral ischemia and reperfusion injury in mice by suppressing oxidative stress,” Stroke, vol. 46, no. 2, pp. 513–519, 2015.

[43] P. Li, M. Shen, F. Gao et al., “An antagonir to microRNA-106b-5p ameliorates cerebral ischemia and reperfusion injury in rats via inhibiting apoptosis and oxidative stress,” Molecular Neurobiology, vol. 54, no. 4, pp. 2901–2921, 2017.

[44] H. Zhao, Z. Tao, R. Wang et al., “MicroRNA-23a-3p attenuates oxidative stress injury in a mouse model of focal cerebral ischemia-reperfusion,” Brain Research, vol. 1592, pp. 65–72, 2014.

[45] Z. Tao, H. Zhao, R. Wang et al., “Neuroprotective effect of microRNA-99a against focal cerebral ischemia-reperfusion injury in mice,” Journal of the Neurological Sciences, vol. 355, no. 1–2, pp. 113–119, 2015.

[46] H. Yi, Y. Huang, F. Yang, W. Lui, S. He, and X. Hu, “MicroRNA-182 aggravates cerebral ischemia injury by targeting inhibitory member of the ASPP family (iASPP),” Archives of Biochemistry and Biophysics, vol. 620, pp. 52–58, 2017.

[47] P. Wang, X. Liang, Y. Lu, X. Zhao, and J. Liang, “MicroRNA-93 downregulation ameliorates cerebral ischemic injury through the Nr2f2/HO-1 defense pathway,” Neurochemical Research, vol. 41, no. 10, pp. 2627–2635, 2016.

[48] M. M. Harraz, S. M. Eacker, X. Wang, T. M. Dawson, and V. L. Dawson, “MicroRNA-223 is neuroprotective by targeting glutamate receptors,” Proceedings of the National Academy of Sciences of the United States of America, vol. 109, no. 46, pp. 18962–18967, 2012.

[49] S. Khanna, C. Rink, R. Ghoorkhanian et al., “Loss of miR-29b following acute ischemic stroke contributes to neuronal cell death and infarct size,” Journal of Cerebral Blood Flow and Metabolism, vol. 33, no. 8, pp. 1197–1206, 2013.

[50] G. Pandi, V. P. Nakka, A. Dhara, A. Roopra, and R. Vetugunti, “MicroRNA miR-29c down-regulation leading to de-repression of its target DNA methyltransferase 3a promotes ischemic brain damage,” PloS One, vol. 8, no. 3, article e58039, 2013.

[51] X. S. Liu, M. Chopp, X. L. Wang et al., “MicroRNA-17-92 cluster mediates the proliferation and survival of neural progenitor cells after stroke,” The Journal of Biological Chemistry, vol. 288, no. 18, pp. 12478–12488, 2013.

[52] Y. Sun, H. Gui, Q. Li et al., “MicroRNA-124 protects neurons against apoptosis in cerebral ischemic stroke,” CNS Neuroscience & Therapeutics, vol. 19, no. 10, pp. 813–819, 2013.

[53] J. Qiu, X. Y. Zhou, X. G. Zhou, R. Cheng, H. Y. Liu, and Y. Li, “Neuroprotective effects of microRNA-210 on hypoxic-ischemic encephalopathy,” BioMed Research International, vol. 2013, Article ID 350419, 5 pages, 2013.

[54] Y. Qu, J. Wu, D. Chen et al., “MiR-139-5p inhibits HGTDP-5 and regulates neuronal apoptosis induced by hypoxia-ischemia in neonatal rats,” Neurobiology of Disease, vol. 63, pp. 184–193, 2014.

[55] J. Ni, X. Wang, S. Chen et al., “MicroRNA let-7c-5p protects against cerebral ischemia injury via mechanisms involving the inhibition of microglia activation,” Brain, Behavior, and Immunity, vol. 49, pp. 75–85, 2015.

[56] Y. Li, L. Mao, Y. Gao, S. Baral, Y. Zhou, and B. Hu, “MicroRNA-107 contributes to post-stroke angiogenesis by targeting Dicer-1,” Scientific Reports, vol. 5, no. 1, article 13316, 2015.
[57] J. Tao, W. Liu, G. Shang et al., “MiR-207/352 regulate lysosomal-associated membrane proteins and enzymes following ischemic stroke,” *Neuroscience*, vol. 305, pp. 1–14, 2015.

[58] F. J. Liu, P. Kaur, D. S. Karolina et al., “MiR-335 regulates Hif-1α to reduce cell death in both mouse cell line and rat ischemic models,” *PLoS One*, vol. 10, no. 6, article e0128432, 2015.

[59] H. Yu, M. Wu, P. Zhao, Y. Huang, W. Wang, and W. Yin, “Neuroprotective effects of viral overexpression of microRNA-22 in rat and cell models of cerebral ischemia-reperfusion injury,” *Journal of Cellular Biochemistry*, vol. 116, no. 2, pp. 233–241, 2015.

[60] N. Wei, L. Xiao, R. Xue et al., “MicroRNA-9 mediates the cell apoptosis by targeting Bcl2H1 in ischemic stroke,” *Molecular Neurobiology*, vol. 53, no. 10, pp. 6809–6817, 2016.

[61] N. Zhang, J. Zhong, S. Han, Y. Li, Y. Yin, and J. Li, “MicroRNA-378 alleviates cerebral ischemic injury by negatively regulating apoptosis executioner caspase-3,” *International Journal of Molecular Sciences*, vol. 17, no. 9, p. 1427, 2016.

[62] D. Z. Liu, G. C. Jickling, B. P. Ander et al., “Elevating microRNA-122 in blood improves outcomes after temporary middle cerebral artery occlusion in rats,” *Journal of Cerebral Blood Flow and Metabolism*, vol. 36, no. 8, pp. 1374–1383, 2016.

[63] L. L. Zeng, X. S. He, J. R. Liu, C. B. Zheng, Y. T. Wang, and G. Y. Yang, “Lentivirus-mediated overexpression of MicroRNA-210 improves long-term outcomes after focal cerebral ischemia in mice,” *CNS Neuroscience & Therapeutics*, vol. 22, no. 12, pp. 961–969, 2016.

[64] S. Yao, B. Tang, G. Li, R. Fan, and F. Cao, “miR-545 inhibits neuronal cell death by targeting TRAF3 in cerebral ischemic stroke,” *Neuropsychiatric Disease and Treatment*, vol. Volume 12, pp. 3083–3092, 2016.

[65] A. Selvamani and F. Sohrabi, “Mir363-3p improves ischemic stroke outcomes in female but not male rats,” *Neurochemistry International*, vol. 107, pp. 168–181, 2017.

[66] A. Dharap, K. Bowen, R. Place, L. C. Li, and R. Vemuganti, “Transient focal ischemia induces extensive temporal changes in rat cerebral microRNAome,” *Journal of Cerebral Blood Flow and Metabolism*, vol. 29, no. 4, pp. 675–687, 2009.

[67] S. Sepramaniam, A. Armugam, K. Y. Lim et al., “MicroRNA 320a functions as a novel endogenous modulator of aquaporins 1 and 4 as well as a potential therapeutic target in cerebral ischemia,” *The Journal of Biological Chemistry*, vol. 285, no. 38, pp. 29223–29230, 2010.

[68] K. J. Yin, Z. Deng, H. Huang et al., “miR-497 regulates neuronal death in mouse brain after transient focal cerebral ischemia,” *Neurobiology of Disease*, vol. 38, no. 1, pp. 17–26, 2010.

[69] A. Selvamani, P. Sathyam, R. C. Miranda, and F. Sohrabi, “An antagonist to microRNA Let7f promotes neuroprotection in an ischemic stroke model,” *PLoS One*, vol. 7, no. 2, article e32662, 2012.

[70] J. M. Moon, L. Xu, and R. G. Giffard, “Inhibition of microRNA-181 reduces forebrain ischemia-induced neuronal loss,” *Journal of Cerebral Blood Flow and Metabolism*, vol. 33, no. 12, pp. 1976–1982, 2013.

[71] Z. Peng, J. Li, Y. Li et al., “Downregulation of miR-181b in mouse brain following ischemic stroke induces neuroprotection against ischemic injury through targeting heat shock protein A5 and ubiquitin carboxyl-terminal hydrolase isozyome L1,” *Journal of Neuroscience Research*, vol. 91, no. 10, pp. 1349–1362, 2013.

[72] A. Vinciguerra, L. Formisano, P. Cerullo et al., “MicroRNA-103-1 selectively downregulates brain NCX1 and its inhibition by anti-miRNA ameliorates stroke damage and neurological deficits,” *Molecular Therapy*, vol. 22, no. 10, pp. 1829–1838, 2014.

[73] P. Wang, J. Liang, Y. Li et al., “Down-regulation of miRNA-30a alleviates cerebral ischemic injury through enhancing beclin 1-mediated autophagy,” *Neurochemical Research*, vol. 39, no. 7, pp. 1279–1291, 2014.

[74] F. Zhu, J. L. Liu, J. P. Li, F. Xiao, Z. X. Zhang, and Z. Zhang, “MicroRNA-124 (mir-124) regulates Ku70 expression and is correlated with neuronal death induced by ischemia/reperfusion,” *Journal of Molecular Neuroscience*, vol. 52, no. 1, pp. 148–155, 2014.

[75] W. Chi, F. Meng, Y. Li et al., “Impact of microRNA-134 on neural cell survival against ischemic injury in primary cultured neuronal cells and mouse brain with ischemic stroke by targeting HSPA12B,” *Brain Research*, vol. 1592, pp. 22–33, 2014.

[76] C. M. Stary, L. Xu, X. Sun et al., “MicroRNA-200c contributes to injury from transient focal cerebral ischemia by targeting Reelin,” *Stroke*, vol. 46, no. 2, pp. 551–556, 2015.

[77] G. Xing, Z. Luo, C. Zhong, X. Pan, and X. Xu, “Influence of mir-155 on cell apoptosis in rats with ischemic stroke: role of the Ras homolog enriched in brain (Rheb)/mTOR pathway,” *Medical Science Monitor*, vol. 22, pp. 5141–5153, 2016.

[78] W. Liu, X. Chen, and Y. Zhang, “Effects of microRNA-21 and microRNA-24 inhibitors on neuronal apoptosis in ischemic stroke,” *American Journal of Translational Research*, vol. 8, no. 7, pp. 3179–3187, 2016.

[79] Q. Li, Q. He, S. Baral et al., “MicroRNA-493 regulates angiogenesis in a rat model of ischemic stroke by targeting MIF,” *The FEBS Journal*, vol. 283, no. 9, pp. 1720–1733, 2016.

[80] L. Pei, S. Meng, W. Yu, Q. Wang, F. Song, and L. Ma, “Inhibition of microRNA-383 ameliorates injury after focal cerebral ischemia via targeting PPARβ,” *Cellular Physiology and Biochemistry*, vol. 39, no. 4, pp. 1339–1346, 2016.

[81] X. Yang, X. Tang, P. Sun et al., “MicroRNA-15a/16-1 antagonist ameliorates ischemic brain injury in experimental stroke,” *Stroke*, vol. 48, no. 7, pp. 1941–1947, 2017.

[82] F. Zhao, Y. Qu, J. Zhu et al., “miR-30d-5p plays an important role in autophagy and apoptosis in developing rat brains after hypoxic-ischemic injury,” *Journal of Neuropathology and Experimental Neurology*, vol. 76, no. 8, pp. 709–719, 2017.

[83] Y. Fan, S. Ding, Y. Sun, B. Zhao, Y. Pan, and J. Wan, “MiR-377 regulates inflammation and angiogenesis in rats after cerebral ischemic injury,” *Journal of Cellular Biochemistry*, vol. 119, no. 1, pp. 327–337, 2018.

[84] X. Wang, Y. Suofu, B. Akpinar et al., “Systemic antimiR-337-3p delivery inhibits cerebral ischemia-mediated injury,” *Neurobiology of Disease*, vol. 105, pp. 156–163, 2017.

[85] S. N. Neifert, E. K. Chapman, M. L. Martini et al., “Aneurysmal subarachnoid hemorrhage: the last decade,” *Translational Stroke Research*, vol. 12, no. 3, pp. 428–446, 2021.

[86] V. L. Feigin, C. M. Lawes, D. A. Bennett, S. L. Barker-Collo, and V. Parag, “Worldwide stroke incidence and early case fatality reported in 56 population-based studies: a systematic review,” *Lancet Neurology*, vol. 8, no. 4, pp. 355–369, 2009.
[87] J. W. Hop, G. J. E. Rinkel, A. Algra, and J. van Gijn, “Case-fatality rates and functional outcome after subarachnoid hemorrhage,” *Stroke*, vol. 28, no. 3, pp. 660–664, 1997.

[88] K. Li, C. D. Barras, R. V. Chandra et al., “A review of the management of cerebral vasospasm after aneurysmal subarachnoid hemorrhage,” *World Neurosurgery*, vol. 126, pp. 513–527, 2019.

[89] E. S. Connolly Jr., A. A. Rabinstein, J. R. Carhuapoma et al., “Guidelines for the management of aneurysmal subarachnoid hemorrhage,” *Stroke*, vol. 43, no. 6, pp. 1711–1737, 2012.

[90] X. W. Su, A. H. Y. Chan, G. Lu et al., “Circulating microRNA 132-3p and 324-3p profiles in patients after acute aneurysmal subarachnoid hemorrhage,” *PLoS One*, vol. 10, no. 12, article e0144724, 2015.

[91] N. S. Lai, J. Q. Zhang, F. Y. Qin, B. Sheng, X. G. Fang, and Z. B. Li, “Serum microRNAs are non-invasive biomarkers for the presence and progression of subarachnoid haemorrhage,” *Bioscience Reports*, vol. 37, no. 1, 2017.

[92] B. Sheng, N. S. Lai, Y. Yao et al., “Early serum miR-1297 is an indicator of poor neurological outcome in patients with aSAH,” *Bioscience Reports*, vol. 38, no. 6, 2018.

[93] C. J. Powers, R.Dickerson, S. W. Zhang, C. Rink, S. Roy, and C. K. Sen, “Human cerebrospinal fluid microRNA: temporal changes following subarachnoid hemorrhage,” *Physiological Genomics*, vol. 48, no. 5, pp. 361–366, 2016.

[94] S. S. Styli, A. A. Adamides, R. M. Koldej et al., “miRNA expression profiling of cerebrospinal fluid in patients with aneurysmal subarachnoid hemorrhage,” *Journal of Neurosurgery*, vol. 126, no. 4, pp. 1131–1139, 2017.

[95] G. Lu, M. S. Wong, M. Z. Q. Xiong et al., “Circulating microRNAs in delayed cerebral infarction after aneurysmal subarachnoid hemorrhage,” *Journal of the American Heart Association*, vol. 6, no. 4, article e005363, 2017.

[96] S. Bache, R. Rasmussen, M. Roosning, F. P. Laigaard, F. C. Nielsen, and K. Møller, “MicroRNA changes in cerebrospinal fluid after subarachnoid hemorrhage,” *Stroke*, vol. 48, no. 9, pp. 2391–2398, 2017.

[97] Y. Kikkawa, T. Ogura, H. Nakajima et al., “Altered expression of microRNA-15a and Kruppel-like factor 4 in cerebrospinal fluid after aneurysmal subarachnoid hemorrhage,” *World Neurosurgery*, vol. 108, pp. 909–916.e3, 2017.

[98] C. Y. Tan, M. Tian, L. L. Zhang et al., “lncRNA signature for predicting cerebral vasospasm in patients with SAH: implications for precision neurosurgery,” *Molecular Therapy-Nucleic Acids*, vol. 21, pp. 983–990, 2020.

[99] N. Lai, D. Wu, T. Liang et al., “Systemic exosomal miR-193b-3p delivery attenuates neuroinflammation in early brain injury after subarachnoid hemorrhage in mice,” *Journal of Neuroinflammation*, vol. 17, no. 1, p. 74, 2020.

[100] L. Wang, Y. Zhao, S. Gang et al., “Inhibition of miR-103-3p preserves neurovascular integrity through caveolin-1 in experimental subarachnoid hemorrhage,” *Neuroscience*, vol. 461, pp. 91–101, 2021.

[101] T. H. Tsai, C. H. Chang, S. H. Lin et al., “Therapeutic effect of and mechanisms underlying the effect of miR-195-5p on subarachnoid hemorrhage-induced vasospasm and brain injury in rats,” *PeerJ*, vol. 9, article e11395, 2021.

[102] X. Gao, Y. Xiong, Q. Li et al., “Extracellular vesicle-mediated transfer of miR-21-5p from mesenchymal stromal cells to neurons alleviates early brain injury to improve cognitive function via the PTEN/Akt pathway after subarachnoid hemorrhage,” *Cell Death & Disease*, vol. 11, no. 5, pp. 1–16, 2020.

[103] J. Li, J. L. Tri, G. Q. Lin, L. F. Xiao, G. L. Su, and L. M. Yang, “lncRNA MEG3 participates in neuronal cell injury induced by subarachnoid hemorrhage via inhibiting the PI3K/Akt pathway,” *European Review for Medical and Pharmacological Sciences*, vol. 22, no. 9, pp. 2824–2831, 2018.

[104] X. Q. Qin, F. Akter, L. Qin et al., “MicroRNA-26b/PTEN signaling pathway mediates glycine-induced neuroprotection in SAH injury,” *Neurochemical Research*, vol. 44, no. 11, pp. 2658–2669, 2019.

[105] X. R. Xu, J. Q. Li et al., “miR-706 alleviates white matter injury via downregulating PKCa/MST1/NF-κB pathway after subarachnoid hemorrhage in mice,” *Experimental Neurology*, vol. 341, article 113688, 2021.

[106] H. Zhao, Y. Li, L. Chen et al., “HucMSCs-derived miR-206-knockdown exosomes contribute to neuroprotection in subarachnoid hemorrhage induced early brain injury by targeting BDNF,” *Neuroscience*, vol. 417, pp. 11–23, 2019.

[107] S. Yang, W. Tang, Y. He, L. Wen, B. Sun, and S. Li, “Long non-coding RNA and microRNA-675/let-7a mediates the protective effect of melatonin against early brain injury after subarachnoid hemorrhage via targeting TP53 and neural growth factor,” *Cell Death & Disease*, vol. 9, no. 2, pp. 199, 2018.

[108] J. Suzuki and A. Takaku, “Cerebrovascular ‘moyamoya’ disease. Disease showing abnormal net-like vessels in base of brain,” *Archives of Neurology*, vol. 20, no. 3, pp. 288–299, 1969.

[109] D. G. Weinberg, R. J. Rahme, S. G. Aoun, H. H. Batjer, and B. R. Bendok, “Moyamoya disease: functional and neurocognitive outcomes in the pediatric and adult populations,” *Neurosurgical Focus*, vol. 30, no. 6, article E21, 2011.

[110] J. R. Festa, L. R. Schwarz, N. Piskin et al., “Neurocognitive dysfunction in adult moyamoya disease,” *Journal of Neurology*, vol. 257, no. 5, pp. 806–815, 2010.

[111] S. Kuriyama, Y. Kusaka, M. Fujimura et al., “Prevalence and clinicoepidemiological features of moyamoya disease in Japan: findings from a nationwide epidemiological survey,” *Stroke*, vol. 39, no. 1, pp. 42–47, 2008.

[112] S. Miyamoto, T. Yoshimoto, N. Hashimoto et al., “Effects of extracranial-intracranial bypass for patients with hemorrhagic moyamoya disease: results of the Japan Adult Moyamoya Trial,” *Stroke*, vol. 45, no. 5, pp. 1415–1421, 2014.

[113] “Guidelines for diagnosis and treatment of moyamoya disease (spontaneous occlusion of the circle of Willis),” *Neurologia Medico-Chirurgica (Tokyo)*, vol. 52, no. 5, pp. 245–266, 2012.

[114] M. Fukui, “Guidelines for the diagnosis and treatment of spontaneous occlusion of the circle of Willis (‘moyamoya’ disease). Research Committee on Spontaneous Occlusion of the Circle of Willis (Moyamoya Disease) of the Ministry of Health and Welfare,” *Clinical Neurology And Neurosurgery*, vol. 99, Suppl 2, pp. S238–S240, 1997.

[115] D. Dai, Q. Lu, Q. Huang et al., “Serum miRNA signature in moyamoya disease,” *PLoS One*, vol. 9, no. 8, article e102382, 2014.

[116] C. Wang, M. Zhao, J. Wang, D. Zhang, S. Wang, and J. Zhao, “Expression analysis of transfer RNA-derived fragments in the blood of patients with moyamoya disease: a preliminary study,” *Molecular Medicine Reports*, vol. 19, no. 5, pp. 3564–3574, 2019.
[117] Q. Ma, L. Li, B. Yu et al., “Circular RNA profiling of neutrophil transcriptome provides insights into asymptomatic moyamoya disease,” *Brain Research*, vol. 1719, pp. 104–112, 2019.

[118] M. Zhao, F. Gao, D. Zhang et al., “Altered expression of circular RNAs in moyamoya disease,” *Journal of the Neurological Sciences*, vol. 381, pp. 25–31, 2017.

[119] G. Wang, Y. Wen, O. D. Faleti et al., “A panel of exosome-derived miRNAs of cerebrospinal fluid for the diagnosis of moyamoya disease,” *Frontiers in Neuroscience*, vol. 14, article 548278, 2020.

[120] X. Chen, Y. Ba, L. Ma et al., “Characterization of microRNAs in serum: a novel class of biomarkers for diagnosis of cancer and other diseases,” *Cell Research*, vol. 18, no. 10, pp. 997–1006, 2008.

[121] C. Wang, B. Gong, P. R. Bushel et al., “The concordance between RNA-seq and microarray data depends on chemical treatment and transcript abundance,” *Nature Biotechnology*, vol. 32, no. 9, pp. 926–932, 2014.

[122] SEQC Consortium, “A comprehensive assessment of RNA-seq accuracy, reproducibility and information content by the Sequencing Quality Control Consortium,” *Nature Biotechnology*, vol. 32, no. 9, pp. 903–914, 2014.

[123] M. Weber, M. B. Baker, R. S. Patel, A. A. Quyyumi, G. Bao, and C. D. Searles, “MicroRNA expression profile in CAD patients and the impact of ACEI/ARB,” *Cardiology Research and Practice*, vol. 2011, Article ID 532915, 5 pages, 2011.

[124] J. N. Boeckel, C. E. Thomé, D. Leistner, A. M. Zeiher, S. Fichtlscherer, and S. Dimmeler, “Heparin selectively affects the quantification of microRNAs in human blood samples,” *Clinical Chemistry*, vol. 59, no. 7, pp. 1125–1127, 2013.

[125] S. Fichtlscherer, S. de Rosa, H. Fox et al., “Circulating microRNAs in patients with coronary artery disease,” *Circulation Research*, vol. 107, no. 5, pp. 677–684, 2010.

[126] H. C. de Boer, C. van Solingen, J. Prins et al., “Aspirin treatment hampers the use of plasma microRNA-126 as a biomarker for the progression of vascular disease,” *European Heart Journal*, vol. 34, no. 44, pp. 3451–3457, 2013.

[127] J. Viereck and T. Thum, “Circulating noncoding RNAs as biomarkers of cardiovascular disease and injury,” *Circulation Research*, vol. 120, no. 2, pp. 381–399, 2017.

[128] V. Mouraviev, B. Lee, V. Patel et al., “Clinical prospects of long noncoding RNAs as novel biomarkers and therapeutic targets in prostate cancer,” *Prostate Cancer and Prostatic Diseases*, vol. 19, no. 1, pp. 14–20, 2016.

[129] P. S. Mitchell, R. K. Parkin, E. M. Kroh et al., “Circulating microRNAs as stable blood-based markers for cancer detection,” *Proceedings of the National Academy of Sciences of the United States of America*, vol. 105, no. 30, pp. 10513–10518, 2008.

[130] J. S. Shah, P. S. Soon, and D. J. Marsh, “Comparison of methodologies to detect low levels of hemolysis in serum for accurate assessment of serum microRNAs,” *PLoS One*, vol. 11, no. 4, article e0153200, 2016.

[131] K. M. Danielson, R. Rubio, F. Abderazzaq, S. Das, and Y. E. Wang, “High throughput sequencing of extracellular RNA from human plasma,” *PLoS One*, vol. 12, no. 1, article e0164644, 2017.