Neurosci. Bull. October, 2016, 32(5):463–468
DOI 10.1007/s12264-016-0038-5

www.neurosci.cn
www.springer.com/12264

REVIEW

Blood Biomarkers in Minor Stroke and Transient Ischemic Attack

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Received: 19 April 2016 / Accepted: 5 May 2016 / Published online: 1 June 2016

Abstract Minor stroke and transient ischemic attack (TIA) are common disorders with a high rate of subsequent disabling stroke, so the early recognition and management of minor stroke and TIA is of great importance. At the moment, the diagnosis of these disorders is based on neurologic deficits in a stroke-clinician's examination of the patient, supplemented by the results of acute brain imaging. However, high variability in TIA diagnosis has been reported between physicians, even trained vascular neurologists, and image-based diagnostic confirmation is not always readily available. Some patients still have ischemic events despite sustained standard secondary preventive therapy. Blood biomarkers are promising to aid in the diagnosis, risk stratification, and individual treatment of minor stroke and TIA. Some studies are being conducted in this field. This mini-review aims to highlight potential biomarkers for diagnosis and those helpful in predicting the risk of future stroke and the selection of treatment.

Keywords Minor stroke · TIA · Biomarker · Prognosis · Diagnosis · Gene · Protein

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Introduction

Epidemiologic studies have shown that minor ischemic strokes account for 30% of annual new strokes, which are defined as strokes with mild and non-disabling symptoms [1, 2]; a recent cross-sectional nationwide study showed that the age-standardized prevalence of TIA is 2.27% in China [3]. Early initiation of preventive therapy after TIA or minor stroke is associated with an 80% reduction in the risk of recurrent stroke within 3 months [4], indicating that early recognition of TIA or minor stroke is of great importance.

At the moment, the diagnosis of ischemic stroke is based on neurologic deficits in a stroke-clinician's examination of the patient, supplemented by the results of acute brain imaging. However, high variability in TIA diagnosis has been reported between physicians, even trained vascular neurologists [5, 6]. The diagnosis of minor stroke has been greatly improved by sophisticated multimodal brain imaging and the use of magnetic resonance imaging (MRI), which has greater sensitivity and specificity in detecting ischemia than traditional imaging tools like computed tomography [7]. However, none of these imaging modalities have achieved a satisfactory diagnostic value. The problems are further aggravated by contraindications for MRI or its unavailability in small hospitals or at night. Thus, these issues call for the development of an easily-accessible test that is available 24-hours per day to further improve the correctness and convenience of diagnosis. Evidence supports the idea that biomarkers play a role in the diagnosis of minor stroke and TIA, and indicates that measurements of biomarkers could be promising. The analogy to troponin for the diagnosis of acute coronary syndromes is obvious.

Minor ischemic stroke or TIA *per se* does not result in major disability, but it has a substantial risk of a subsequent ischemic stroke. It has been estimated that after the index

event, 10 to 20% of these patients have a stroke within 3 months, most of which occur within the first 2 days [8–11]. A recent study reported a lower risk of recurrent stroke due to better implementation of secondary prevention as recommended by current guidelines [12]. However, the prognosis of recurrent stroke is mostly unfavorable [13]. Several clinical conventional risk factors and higher $ABCD^2$ scores have been associated with recurrent stroke, but these factors do not fully explain the risk of recurrent stroke [12, 14].

In addition to the conventional risk factors of recurrent stroke, researchers have identified several biomarkers to predict the occurrence and progression of minor stroke and TIA. These biomarkers play a critical role in confirming the ischemic etiology of transient or mild neurological deficits and may serve as a surrogate endpoint in guiding the choice of management strategy. In this review, we first elaborate on potential biomarkers for the diagnosis of minor stroke and TIA, and then focus on those that may help in predicting the risk of future stroke and guiding the choice of treatment.

Biomarkers in Diagnosis

Genes

Plasminogen Activator Inhibitor 1

Genetic risk factors play an important role in the etiology of vascular diseases. The insertion/deletion polymorphism 4G/5G in the promoter region of the plasminogen activator inhibitor 1 (PAI-1) gene has been associated with an increased risk of myocardial infarction [15, 16]. A prior study found that the 4G/4G genotype tends to be less frequent in patient with minor stoke and TIA, especially in young patients [17].

mtDNA Sub-haplogroup K

Chinnery *et al.* investigated the mtDNA haplogroup distribution in two independent UK cohorts of patients with TIA or ischemic stroke and found that genetic variation of mtDNA sub-haplogroup K is an independent determinant of the risk of cerebral, but not coronary, ischemic vascular events. These findings suggest mitochondrial mechanisms in the etiology of ischemic stroke and provide a new approach to identifying individuals with a high susceptibility to developing ischemic stroke [18].

5,10-Methylenetetrahydrofolate Reductase

Homocysteine might be a potential predictor of ischemic stroke or TIA. A common C677T mutation in the gene for 5,10-methylenetetrahydrofolate reductase (5,10-MTHFR) has been linked to elevated homocysteine levels and has therefore been suspected to be a candidate genetic risk factor for arterial occlusive disease. The authors investigated the prevalence of the C677T MTHFR mutation in addition to factor V Leiden mutation in patients with minor stroke or TIA. No differences in the prevalence of the MTHFR mutation were found between patients and controls, whereas there was a trend towards a higher prevalence in carriers of factor V Leiden in the patients. The combination of C677T MTHFR mutation and factor V Leiden might unfavorably affect the clinical course [19].

Proteins

Platelet Basic Protein

Besides genetic methods, the emerging science of proteomics provides a unique platform for investigating biomarkers of ischemic disease. A recent study with a small sample size used mass spectroscopy-based proteomics to identify serum biomarkers of minor stroke and TIA. By verification in a second cohort of patients, platelet basic protein was identified as a candidate serum biomarker for TIA [20].

Biomarkers in Stroke Subtypes

Protein

Homocysteine Ischemic stroke is a heterogeneous disease with variable pathogenesis [21]. Homocysteine has been shown to cause vascular damage by damaging the vascular matrix and augmenting the proliferation of vascular smooth muscle [22]. An association between increased plasma homocysteine and ischemic stroke caused by large-artery atherosclerosis and small-artery disease was reported in earlier studies [23, 24]. A previous study then investigated the relationship between serum homocysteine levels and ischemic stroke subtypes in patients with acute ischemic stroke within 7 days of symptom onset or TIA. The homocysteine level was measured on the second day after admission. They found that, among the subtypes of ischemic stroke, patients with small-vessel disease and confluent leukoaraiosis had the highest homocysteine levels [25].

Biomarkers in Prognosis

Proteins

Unlike the diagnostic biomarkers, those helpful in risk prediction are less likely to come from injured brain tissue,

but rather from the pathological substrate underlying arterial occlusion, which in most cases would be an atherosclerotic plaque. Therefore, an active plaque that is at high risk for rupture and thrombus formation should be the main focus of the search for biomarkers predicting the risk of future vascular events. This could further help to identify patients most likely to benefit from endovascular therapy or surgery. Some of the biomarkers aimed at the prognosis of minor stroke or TIA are summarized in Table 1.

Lipoprotein-Associated Phospholipase A2 and Soluble CD40L

Both lipoprotein-associated phospholipase A2 (Lp-PLA2) and soluble CD40 ligand (sCD40L) have been linked to atherosclerotic plaque inflammation and instability [26, 27]. The result of the Clopidogrel in High-Risk Patients With Acute Non-disabling Cerebrovascular Events (CHANCE) trial indicated that Clopidogrel plus aspirin is superior to aspirin alone in reducing the risk of recurrent stroke among patients with acute minor stroke and TIA [28]. Two sub-studies of the CHANCE trial focusing on blood biomarkers in prognosis have been reported recently. They included ~3000 samples [29, 30]. The authors determined the levels of Lp-PLA2 and sCD40L in the acute period and found that both were associated with an increased risk of recurrent vascular events within a 90-day follow-up.

High-Sensitive C-Reactive Protein

hsCRP is one of the most-investigated inflammatory markers for the prognosis of ischemic events. It has also been associated with active plaques. hsCRP levels determined within 24 h of symptom onset in patients with TIA have been associated with an increased risk of recurrent stroke [31].

On the other hand, several studies have elaborated the predictive value of hsCRP in mortality, but few have investigated its association with functional disability in patients with stroke. A recent study recruited 977 patients with acute non-cardioembolic ischemic stroke or TIA with middle cerebral artery stenosis, and found that elevated hs-CRP levels independently predicted a 1-year poor outcome defined as Modified Rankin Scale (mRS) 3–6 [32]. However, the patients with major stroke were also included in this study.

Antiphospholipid Antibodies

Antiphospholipid antibodies are a diverse collection of autoantibodies which are directed at plasma proteins bound to phospholipid membranes. These antibodies have been

Table 1 Blood biomarkers in the prognosis of minor stroke and TIA.

Biomarker	Description	References	Potential clinical application	Size	Study design	Time point
Lp-PLA2	Enzyme associated with metabolism of low-density lipoprotein	Lin *et al.* [29]	Risk of ischemic stroke, myocardial infarction, or death	3021 minor stroke and TIA patients	Observational study nested in an RCT trial	<48 h after symptom onset
sCD40L	Soluble CD40L, a member of the tumor necrosis factor family of ligands expressed on CD4+ T cells and activated platelets	Li *et al.* [30]	Risk of recurrent stroke	3044 minor stroke and TIA patients	Observational study nested in an RCT trial	<48 h after symptom onset
hsCRP	Nonspecific systemic inflammation marker	Purroy *et al.* [31], Gong *et al.* [32]	Risk of recurrent stroke; poor neurological outcome (mRS 3-6)	135 TIA patients; 977 ischemic stroke and TIA patients	Cohort study	<24 h after symptom onset; <7 days from symptom onset
aPS/PT IgG	Antiphospholipid antibodies	Mullen *et al.* [35]	Stroke or death	167 TIA patients	Cohort study	<48 h from symptom onset
sTNFR-1, vWF, HFABP and NT-proBNP	Cytokine related to inflammation, thrombosis, and cardiac/neuronal injury	Greisenegger *et al.* [36]	All-cause death, vascular death, cancer death	493 minor stroke and 436 TIA patients	Cohort study	5 days from onset of index event on average

Lp-PLA2 lipoprotein-associated phospholipase A2, *sCD40L* soluble CD40 ligand, *hsCRP* high-sensitivity C-reactive protein, *sTNFR-1* soluble tumor necrosis factor α receptor-1, *vWF* von Willebrand factor, *HFABP* heart-type fatty-acid-binding protein, *NT-proBNP* N-terminal pro-B-type natriuretic peptide.

associated with thrombosis in antiphospholipid antibody syndrome [33] and atherosclerotic vascular disease in individuals without antiphospholipid antibody syndrome [34]. A prior study enrolled 167 patients with TIA within 48 h of symptom onset and measured the levels of antiphospholipid antibodies. They found that anti-phosphatidylserine/prothrombin IgG antibodies are independently associated with stroke or death in patients with TIA [35].

Soluble Tumor Necrosis Factor α receptor-1, von Willebrand Factor, Heart-Type Fatty-Acid-Binding Protein, and N-terminal pro-B-type Natriuretic Peptide

Some blood biomarkers have been found to be predictors of all-cause death, despite the absence of an association with the risk of nonfatal stroke. Greisenegger and colleagues measured fifteen biomarkers in 929 patients with minor stroke and TIA in a population-based study. During 11-years of follow-up, none of the biomarkers predicted the risk of non-fatal vascular events. However, soluble tumor necrosis factor α receptor-1, von Willebrand factor, heart-type fatty-acid-binding protein, and N-terminal pro-B-type natriuretic peptide independently predicted all-cause death. Among these four biomarkers, N-terminal pro-B-type natriuretic peptide was most predictive of vascular death, whereas heart-type fatty-acid-binding protein predicted cancer death. These results might help to improve the selection of patients for additional screening for occult cardiac disease or cancer [36].

Biomarkers of Response to Therapy

Proteins

Though sets of biomarkers to help in the diagnosis and prognosis would be very helpful, this is of little importance compared to the practical question – how to reduce the high risk of recurrence – since the challenge will remain even if we can accurately identify patients with a high risk of future strokes.

Antiplatelet treatment is the mainstay of secondary prevention of ischemic stroke and TIA. However, some patients still have ischemic events despite sustained antiplatelet therapy. Besides clinical risk factors, some markers, such as hsCRP [37, 38], sCD40L [39, 40], fibrinogen [41], and genetic polymorphisms of glycoprotein IIIa, COX-1, COX-2, and P2Y12 [42] have been suggested to be predictive of such failure of antiplatelet treatment in patients with cardiovascular disease. However, there is still controversy with regard to patients with ischemic stroke and little is known about biomarkers for individualized antiplatelet treatment in patients with minor stroke and TIA [43].

Glycated Albumin

The result of another sub-study of the CHANCE trial showed an interaction of baseline glycated albumin (GA) levels with dual and single antiplatelet therapy groups in the CHANCE trial cohort, indicating that GA predicts the effects of dual and single antiplatelet therapy in patients with minor stroke or TIA [44]. This finding might be useful for identifying those minor strokes and TIAs most likely to benefit from dual antiplatelet therapy (those with low GA) and prevent an unnecessary risk of hemorrhage in those unlikely to benefit (those with high GA) [45].

Conclusion and Future Perspectives

Biomarker guidance is a promising strategy to aid in the diagnosis and prognosis of minor stroke and TIA, thereby improving early management. However, many theoretical and practical challenges still stand in the way. First, consensus on the definition of a "minor stroke" is needed. A prior study suggested a definition of conscious patients scoring $\leq$1 on every NIHSS item or patients with NIHSS $\leq$3 is most suitable to define a "minor stroke" for clinical and research purposes, because it gave the best short-term and medium-term outcomes compared with other definitions [46]. Second, most of the recent studies that focused on biomarkers did not consider minor and major stroke as distinct entities. Whether minor stroke and TIA have specific biomarker patterns needs to be confirmed. Third, emphasis should be put on careful and standardized phenotyping of ischemic stroke and stroke subtypes when looking for biomarkers, given that meta-analyses of extant genome-wide data illustrate the need to focus on subtypes of ischemic stroke for gene discovery [47]. Fourth, multi-omics analysis provides a good platform for studies of biomarkers. Epigenetic analysis and gene profiling are used as diagnostic tools in cardiovascular disease and cancer. Similar research tools are now beginning to be used for the diagnosis of stroke. For example, changes in the KCNQ1 methylation pattern have recently been shown to be useful as potential stroke biomarkers [48]. Further studies are needed to determine the role of DNA methylation in the diagnosis and prognosis of minor stroke and TIA. Finally, standardized clinical trials using an accepted protocol for measuring biomarkers and intervention targeting at the biomarkers are needed to test their usefulness.

References

1. Zhao D, Liu J, Wang W, Zeng Z, Cheng J, Liu J, *et al.* Epidemiological transition of stroke in China: twenty-one-year observational study from the Sino-MONICA-Beijing Project. Stroke 2008, 39: 1668–1674.
2. Wang YL, Wu D, Liao X, Zhang W, Zhao X, Wang YJ. Burden of stroke in China. Int J Stroke 2007, 2: 211–213.
3. Wang Y, Zhao X, Jiang Y, Li H, Wang L, Johnston SC, *et al.* Prevalence, knowledge, and treatment of transient ischemic attacks in China. Neurology 2015, 84: 2354–2361.
4. Rothwell PM, Giles MF, Chandratheva A, Marquardt L, Geraghty O, Redgrave JN, *et al.* Effect of urgent treatment of transient ischaemic attack and minor stroke on early recurrent stroke (EXPRESS study): a prospective population-based sequential comparison. Lancet 2007, 370: 1432–1442.
5. Castle J, Mlynash M, Lee K, Caulfield AF, Wolford C, Kemp S, *et al.* Agreement regarding diagnosis of transient ischemic attack fairly low among stroke-trained neurologists. Stroke 2010, 41: 1367–1370.
6. Prabhakaran S, Silver AJ, Warrior L, McClenathan B, Lee VH. Misdiagnosis of transient ischemic attacks in the emergency room. Cerebrovasc Dis 2008, 26: 630–635.
7. Chalela JA, Kidwell CS, Nentwich LM, Luby M, Butman JA, Demchuk AM, *et al.* Magnetic resonance imaging and computed tomography in emergency assessment of patients with suspected acute stroke: a prospective comparison. Lancet 2007, 369: 293–298.
8. Kleindorfer D, Panagos P, Pancioli A, Khoury J, Kissela B, Woo D, *et al.* Incidence and short-term prognosis of transient ischemic attack in a population-based study. Stroke 2005, 36: 720–723.
9. Coull AJ, Lovett JK, Rothwell PM, Oxford Vascular S. Population based study of early risk of stroke after transient ischaemic attack or minor stroke: implications for public education and organisation of services. BMJ 2004, 328: 326.
10. Johnston SC, Gress DR, Browner WS, Sidney S. Short-term prognosis after emergency department diagnosis of TIA. JAMA 2000, 284: 2901–2906.
11. Lovett JK, Coull AJ, Rothwell PM. Early risk of recurrence by subtype of ischemic stroke in population-based incidence studies. Neurology 2004, 62: 569–573.
12. Amarenco P, Lavallee PC, Labreuche J, Albers GW, Bornstein NM, Canhao P, *et al.* One-Year Risk of Stroke after Transient Ischemic Attack or Minor Stroke. N Engl J Med 2016, 374: 1533–1542.
13. Jorgensen HS, Nakayama H, Reith J, Raaschou HO, Olsen TS. Stroke recurrence: predictors, severity, and prognosis. The Copenhagen Stroke Study. Neurology 1997, 48: 891–895.
14. Pepine CJ. Residual risk for secondary ischemic events in patients with atherothrombotic disease: opportunity for future improvements in patient care. Ann Med 2010, 42: 19–35.
15. Onalan O, Balta G, Oto A, Kabakci G, Tokgozoglu L, Aytemir K, *et al.* Plasminogen activator inhibitor-1 4G4G genotype is associated with myocardial infarction but not with stable coronary artery disease. J Thromb Thrombolysis 2008, 26: 211–217.
16. Rallidis LS, Gialeraki A, Merkouri E, Liakos G, Dagres N, Sionis D, *et al.* Reduced carriership of 4G allele of plasminogen activator inhibitor-1 4G/5G polymorphism in very young survivors of myocardial infarction. J Thromb Thrombolysis 2010, 29: 497–502.
17. Endler G, Lalouschek W, Exner M, Mitterbauer G, Haring D, Mannhalter C. The 4G/4G genotype at nucleotide position -675 in the promotor region of the PAI-1 gene is less frequent in young patients with minor stroke than in controls. Br J Haematol 2000, 110: 469–471.
18. Chinnery PF, Elliott HR, Syed A, Rothwell PM. Mitochondrial DNA haplogroups and risk of transient ischaemic attack and ischaemic stroke: a genetic association study. Lancet Neurol 2010, 9: 498–503.
19. Lalouschek W, Aull S, Serles W, Schnider P, Mannhalter C, Pabinger-Fasching I, *et al.* C677T MTHFR mutation and factor V Leiden mutation in patients with TIA/minor stroke: a case-control study. Thromb Res 1999, 93: 61–69.
20. George PM, Mlynash M, Adams CM, Kuo CJ, Albers GW, Olivot JM. Novel TIA biomarkers identified by mass spectrometry-based proteomics. Int J Stroke 2015, 10: 1204–1211.
21. Schulz UG, Flossmann E, Rothwell PM. Heritability of ischemic stroke in relation to age, vascular risk factors, and subtypes of incident stroke in population-based studies. Stroke 2004, 35: 819–824.
22. Hankey GJ, Eikelboom JW. Homocysteine and stroke. Curr Opin Neurol 2001, 14: 95–102.
23. Eikelboom JW, Hankey GJ, Anand SS, Lofthouse E, Staples N, Baker RI. Association between high homocyst(e)ine and ischemic stroke due to large- and small-artery disease but not other etiologic subtypes of ischemic stroke. Stroke 2000, 31: 1069–1075.
24. Khan U, Crossley C, Kalra L, Rudd A, Wolfe CD, Collinson P, *et al.* Homocysteine and its relationship to stroke subtypes in a UK black population: the south London ethnicity and stroke study. Stroke 2008, 39: 2943–2949.
25. Ma Y, Zhao X, Zhang W, Liu L, Wang Y, Fang R. Homocysteine and ischemic stroke subtype: a relationship study in Chinese patients. Neurol Res 2010, 32: 636–641.
26. Wilensky RL, Shi Y, Mohler ER, 3rd, Hamamdzic D, Burgert ME, Li J, *et al.* Inhibition of lipoprotein-associated phospholipase A2 reduces complex coronary atherosclerotic plaque development. Nat Med 2008, 14: 1059–1066.
27. Schonbeck U, Libby P. CD40 signaling and plaque instability. Circ Res 2001, 89: 1092–1103.
28. Wang Y, Zhao X, Liu L, Wang D, Wang C, Li H, *et al.* Clopidogrel with aspirin in acute minor stroke or transient ischemic attack. N Engl J Med 2013, 369: 11–19.
29. Lin J, Zheng H, Cucchiara BL, Li J, Zhao X, Liang X, *et al.* Association of Lp-PLA2-A and early recurrence of vascular events after TIA and minor stroke. Neurology 2015, 85: 1585–1591.
30. Li J, Wang Y, Lin J, Wang D, Wang A, Zhao X, *et al.* Soluble CD40L Is a Useful Marker to Predict Future Strokes in Patients With Minor Stroke and Transient Ischemic Attack. Stroke 2015, 46: 1990–1992.
31. Purroy F, Montaner J, Molina CA, Delgado P, Arenillas JF, Chacon P, *et al.* C-reactive protein predicts further ischemic events in transient ischemic attack patients. Acta Neurol Scand 2007, 115: 60–66.
32. Gong X, Zou X, Liu L, Pu Y, Wang Y, Pan Y, *et al.* Prognostic value of inflammatory mediators in 1-year outcome of acute ischemic stroke with middle cerebral artery stenosis. Mediators Inflamm 2013, 2013: 850714.
33. Cohen D, Berger SP, Steup-Beekman GM, Bloemenkamp KW, Bajema IM. Diagnosis and management of the antiphospholipid syndrome. BMJ 2010, 340: c2541.
34. Brey RL, Abbott RD, Curb JD, Sharp DS, Ross GW, Stallworth CL, *et al.* beta(2)-Glycoprotein 1-dependent anticardiolipin antibodies and risk of ischemic stroke and myocardial infarction: the honolulu heart program. Stroke 2001, 32: 1701–1706.
35. Mullen MT, Messe SR, Kasner SE, Sansing L, Husain MR, Norman GL, *et al.* Anti-Phosphatidylserine-Prothrombin Antibodies are Associated with Outcome in a TIA Cohort. Front Neurol 2012, 3: 137.

36. Greisenegger S, Segal HC, Burgess AI, Poole DL, Mehta Z, Rothwell PM. Biomarkers and mortality after transient ischemic attack and minor ischemic stroke: population-based study. Stroke 2015, 46: 659–666.
37. Park DW, Lee SW, Yun SC, Song HG, Ahn JM, Lee JY, *et al.* A point-of-care platelet function assay and C-reactive protein for prediction of major cardiovascular events after drug-eluting stent implantation. J Am Coll Cardiol 2011, 58: 2630–2639.
38. Park DW, Yun SC, Lee JY, Kim WJ, Kang SJ, Lee SW, *et al.* C-reactive protein and the risk of stent thrombosis and cardiovascular events after drug-eluting stent implantation. Circulation 2009, 120: 1987–1995.
39. Ge H, Zhou Y, Liu X, Nie X, Wang Z, Guo Y, *et al.* Relationship between plasma inflammatory markers and platelet aggregation in patients with clopidogrel resistance after angioplasty. Angiology 2012, 63: 62–66.
40. Heeschen C, Dimmeler S, Hamm CW, van den Brand MJ, Boersma E, Zeiher AM, *et al.* Soluble CD40 ligand in acute coronary syndromes. N Engl J Med 2003, 348: 1104–1111.
41. Feher G, Koltai K, Papp E, Alkonyi B, Solyom A, Kenyeres P, *et al.* Aspirin resistance: possible roles of cardiovascular risk factors, previous disease history, concomitant medications and haemorrheological variables. Drugs Aging 2006, 23: 559–567.
42. Topcuoglu MA, Arsava EM, Ay H. Antiplatelet resistance in stroke. Expert Rev Neurother 2011, 11: 251–263.
43. Lai PT, Chen SY, Lee YS, Ho YP, Chiang YY, Hsu HY. Relationship between acute stroke outcome, aspirin resistance, and humoral factors. J Chin Med Assoc 2012, 75: 513–518.
44. Li J, Wang Y, Wang D, Lin J, Wang A, Zhao X, *et al.* Glycated albumin predicts the effect of dual and single antiplatelet therapy on recurrent stroke. Neurology 2015, 84: 1330–1336.
45. Jickling GC. Comment: TIA response to antiplatelets stratified by glycated albumin. Neurology 2015, 84: 1334.
46. Fischer U, Baumgartner A, Arnold M, Nedeltchev K, Gralla J, De Marchis GM, *et al.* What is a minor stroke? Stroke 2010, 41: 661–666.
47. Meschia JF, Arnett DK, Ay H, Brown RD, Jr., Benavente OR, Cole JW, *et al.* Stroke Genetics Network (SiGN) study: design and rationale for a genome-wide association study of ischemic stroke subtypes. Stroke 2013, 44: 2694–2702.
48. Gomez-Uriz AM, Milagro FI, Mansego ML, Cordero P, Abete I, De Arce A, *et al.* Obesity and ischemic stroke modulate the methylation levels of KCNQ1 in white blood cells. Hum Mol Genet 2015, 24: 1432–1440.