ORIGINAL RESEARCH

Correlation of Serum β2-MG, HGF, Lp-PLA2 with Carotid Atherosclerosis in Patients with Hypertension Combined with Cerebral Infarction and their Prognostic Value

Jinhua Wang, MM; Tingting Duan, MM; Zhizhi Chen, MM; Feifei Shao, MM; Ming Yang, MM; Jiao Wang, MD

ABSTRACT

Cerebral infarction is characterized by a high morbidity, disability, and fatality rate. This study explored the relationship between serum β2 microglobulin (β2-MG), HGF, lipoprotein-associated phospholipase A2 (Lp-PLA2) and carotid atherosclerosis in patients with hypertension combined with cerebral infarction and their prognostic value. A total of 320 patients with cerebral infarction complicated with hypertension who were hospitalized from January 2015 to January 2020 were collected. HGF, Lp-PLA2 and β2-MG levels were detected. Plaque score (Crouse score) was the patient's cumulative plaque thickness measurements. Additionally, the maximum plaque thickness and the number of plaques were measured.. The correlation was found between high β2-MG levels and the poor prognosis (HR: 1.29, 95% CI: 1.03-1.52, P = .022). Patients who had elevated levels of HGF were also less likely to have a positive outcome (HR: 1.38, 95% CI: 1.26-1.56, P = .015). High Lp-PLA2 levels were associated with a worse prognosis than low levels (HR: 1.74, 95% CI: 1.29-2.32, P = .015). In conclusion, the levels of β2-MG, HGF, and Lp-PLA2 in patients with hypertension combined with cerebral infarction were substantially linked with carotid plaques. (*Altern Ther Health Med.* [E-pub ahead of print.])

Jinhua Wang, MM; **Tingting Duan,** MM; **Zhizhi Chen,** MM; **Feifei Shao,** MM; **Ming Yang,** MM; Department of Neurology, The Quzhou Affiliated Hospital of Wenzhou Medical University (Quzhou People's Hospital), Quzhou, Zhejiang, China. **Jiao Wang,** MD, Department of Cardiology, The Quzhou Affiliated Hospital of Wenzhou Medical University (Quzhou People's Hospital), Quzhou, Zhejiang, China.

Corresponding author: Jiao Wang, MD
E-mail: wuw1911@163.com

INTRODUCTION

Cerebral infarction is characterized by a high morbidity, disability, and fatality rate, and it brings a serious economic burden to society and families. Large arterial atherosclerosis (LAA), small artery occlusion (SAO), cardiogenic cerebral embolism (CCE), other reasons, and unknown causes (etc.) are the five main subtypes of cerebral infarction according to the widely accepted TOAST classification. The LAA type and SAO type are the most common in clinical practice. The LAA type is an arge-area infarction caused by atherosclerosis of the large arteries. In contrast, the SAO type is a small lacunar infarction caused by the lesions of the perforating arteries or the terminal arterioles themselves.[1-5] Both are caused by blood vessel blockage in the brain and have many of the same risk factors, including high blood pressure, diabetes, high cholesterol, excessive alcohol use, etc. The current clinical treatments are basically anti-platelet aggregation, anticoagulation, lipid regulation, plaque stabilization, promoting circulation, etc. Imaging studies have shown that patients with SAO type are mostly accompanied by large atherosclerosis, and patients with LAA type also have some lacunar infarcts. Therefore, finding the cause or related pathogenic factors for targeted treatment is particularly important. Although the prognosis for cerebral infarction is often better than that of cerebral hemorrhage, it is terrible for those suffering from a severe condition. The presence of atherosclerotic plaques is a significant clinical risk factor that influences patient outcomes. However, there is currently a shortage of studies examining the clinical variables and prognostic factors associated with carotid atherosclerosis.

Endothelial cells are the source of the multifunctional cytokine hepatocyte growth factor (HGF), also known as the spreading factor (SF). HGF is widely distributed in cells of various organs and tissues and can stimulate the differentiation, proliferation, regeneration, movement, migration, and morphogenesis of various cells, including mitosis, hematopoiesis, cardiac hypertrophy, angiogenesis, and anti-apoptosis. These biological effects are closely related to cardiovascular system diseases.[6] Serum levels of this protein have been reported to be significantly elevated in

individuals with hypertension, acute coronary syndrome, and myocardial infarction in recent years. However, data connecting its concentrations to atherosclerosis in the carotid artery are still insufficient. Recent years have discovered an inflammatory marker linked to cerebral infarction called lipoprotein-associated phospholipase A2 (Lp-PLA2). It can hydrolyze platelet-activating factor, bind to LDL, hydrolyze oxidized smear lipids on LDL, produce new pro-inflammatory substances, and promote the formation of atherosclerosis; it is synthesized and secreted by mature macrophages, T-lymphocytes, and mast cells; and it is regulated by inflammatory substances.[7] The concentration of β2-MG is an independent predictor of cardiovascular and cerebrovascular events in recent years.[8] Since the predictive significance of serum β2-MG, HGF, Lp-PLA2, and carotid atherosclerosis in hypertensive patients with cerebral infarction is unknown, this study set out to investigate these relationships.

METHODS

Patients

A total of 320 patients with cerebral infarction complicated with hypertension who were hospitalized from January 2015 to January 2020 were collected. After admission, they were divided into two groups: LAA type and SAO type by TOAST classification; 200 of them were assigned to the LAA group and 120 to the SAO group. LAA type includes Occlusion or stenosis of internal and external large arteries ≥50%, mainly confirmed by vascular imaging, and at least one or more risk factors or other evidence of atherosclerosis. Exclusion criteria: 1. Cardiogenic cerebral infarction caused by cardiogenic or moderate risk factors; 2. Cerebral infarction lesions other than arterial blood supply due to stenosis ≥50% or complete occlusion of large cerebral arteries.

SAO type includes: 1. There are obvious clinical manifestations of lacunar cerebral infarction, and the lesions with clear boundary on imaging (maximum diameter < 1.5 cm) are consistent with the neurological deficit; 2. Clinical manifestations without obvious lacunar cerebral infarction Symptoms and imaging lesions are small (diameter < 1.5 cm); 3. There are symptoms of lacunar infarction, but it is not obvious, imaging does not detect obvious matching lesions, and further related examinations need to be completed. If the patient has had high blood pressure and diabetes in the past, it can be used as a supportive condition. Exclusion criteria: 1. Cardiac, cerebral infarction or lacunar cerebral infarction caused by intracranial and intracranial large artery disease; 2. Infarct lesions greater than or equal to 1.5 cm below the cortex and cortex. This study has been approved by the ethics committee of the Quzhou-affiliated hospital of Wenzhou medical university (NO.20220815), and informed consent was obtained from all patients.

Laboratory test

Fasting for more than 12 hours prior to admission, 5 ml of cubital venous blood was drawn, and the patient's serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), serum creatinine, blood urea nitrogen, and triglycerides were measured using an automatic biochemical analyzer and reagents. In order to measure Lp-PLA2 and HGF levels, blood samples were taken and analyzed for various lipids and proteins using enzyme-linked immunosorbent assay (ELISA). Measure the patient's urine output first thing in the morning. Urinary β2-MG levels were measured using first-thing-in-the-morning samples.

Carotid plaque measurement

The carotid bifurcation, internal carotid artery, external carotid artery, and bilateral common carotid artery plaque thicknesses were all measured by the Philips EPIQ 5 color Doppler ultrasound machine. The plaque was a distinct echogenic object that stuck out into the lumen and measured greater than 1.3 mm in thickness. The patient's total plaque score (Cruse score) was calculated by adding up the measurements of all of their plaques. Maximum plaque thickness and plaque count were also recorded.

Outcome

The outcome of this study was set as readmission or death. Follow-up began after the patient was discharged from the hospital and was followed up every three months. Patient readmissions or deaths were defined as events. The dates on which events occurred were recorded.

Statistical analysis

Statistical testing was executed using SPSS (version 25.0; IBM; Armonk, NY, USA) and R (version 4.0.5). Mean SD or median (range) was used to describe continuous values. The 2 or Fisher's exact test for categorical data and the student *t* test for continuous values, as applicable, were used to examine differences between groups. Clinical variables linked with prognosis were investigated using Cox regression analysis. Statistically significant results from univariate analyses will be included in the multivariate analyses. $P < .05$ was considered to be significant.

RESULTS

Patients

The patient's baseline data are listed in Table 1. The average patient's age was 60.4. More than female patients, male patients accounted for 65.6% of the total patients. 56.6% of the patients had a history of smoking and 37.8% had a history of alcohol consumption. There were no significant differences between the LAA and SAO groups in their initial conditions. The levels of β2-MG, HGF, and Lp-PLA2 did not differ significantly between the two groups. This study revealed that the SAO group had substantially higher levels of cholesterol and HDL-C than the LAA group.

The association between the β2-MG, HGF, Lp-PLA2 levels with carotid plaque

We next explored the association between the β2-MG, HGF, and Lp-PLA2 levels with the carotid plaque crouse

Table 1. Clinical characteristics of patients

Characteristics	All patients	LAA	SAO	*P* value
n	320	200	120	
Age (years)	60.4±12.7	65.2±10.8	65.3±10.8	.830
Male sex, n (%)	210 (65.6)	130 (65.0)	80 (66.6)	.544
Diabetes mellitus, n (%)	124 (38.7)	78 (39.0)	46 (38.3)	.926
Chronic heart disease, n (%)	184 (57.5)	110 (55.0)	74 (61.7)	.359
Smoker, n (%)	181 (56.6)	108 (54.0)	73 (60.8)	.183
Drinking, n (%)	121 (37.8)	71 (35.5)	50 (41.7)	.224
ALT (IU/L)	18.5±7.4	17.5±6.1	19.2±7.7	.456
AST (IU/L)	22.5±1.8	23.4±3.1	21.8±1.9	.207
Albumin (g/L)	45.3±6.8	43.5±7.5	46.0±6.3	.054
Total bilirubin (μmol/L)	15.3 (9.7-32.2)	15.4 (9.2-33.0)	15.6 (9.2-28.2)	.518
Platelet (×1,000/mm^3)	163.0 (124.0-257.5)	159.0 (109.0-263.0)	169.0 (131.5-223.0)	.906
Triglycerides (mmol/L)	1.0 (0.7-1.3)	1.1 (0.8-1.4)	0.9 (0.7-1.1)	.057
Cholesterol (mmol/L)	4.1 (3.5-4.8)	3.8 (3.0-4.3)	4.5 (3.7-4.9)	<.001
HDL-C (mmol/L)	1.2 (0.9-1.5)	0.8 (0.7-1.0)	1.4 (1.2-1.7)	<.001
LDL-C (mmol/L)	2.4 (1.8-2.9)	2.3 (1.8-2.9)	2.5 (1.9-2.9)	.488
FBS (mmol/L)	5.5 (5.0-6.6)	5.8 (5.1-7.2)	5.5 (5.0-6.6)	.267
Hemoglobin A_{1c} (%)	5.8 (5.2-6.6)	5.9 (5.4-6.7)	5.9 (5.3-7.0)	.901
Creatinine (μmol/L)	77 (65-91)	76 (67-91)	81 (69-95)	.233
C-reactive protein (mg/dL)	4.6 (2.0-9.0)	3.3 (1.3-9.7)	4.7 (2.3-9.0)	.230
Blood urea nitrogen (mmol/L)	5.7±2.3	5.5±1.3	5.9±2.4	.360
β2-MG (ng/L)	2331.1±230.2	2320.2±184.3	2384.6±312.2	.566
HGF (pg/mL)	310.5±23.2	304.8±18.3	315.9±14.6	.489
Lp-PLA2 (μg/L)	190.8±11.2	185.6±5.8	197.3±11.9	.543

Note: Continuous variables are expressed as mean, standard deviation or median (interquartile range) when appropriate.

score/carotid plaque thickness. The β2-MG level was positively associated with the crouse score; the results are presented in Figure 1A. As the results in Figure 1B, the β2-MG level was positively associated with the carotid plaque thickness ($P < .01$). We additionally explored the association between the HGF, Lp-PLA2 levels, and crouse score/carotid plaque thickness. We found similar results, and the results were presented in Figures 2 and 3.

Then we changed the crouse score and carotid plaque thickness into the categorical variables. The cutoff for the crouse score was set as 15, and the cutoff for the carotid plaque thickness was set as 2 mm. Clinical variables influencing the Crouse score/carotid plaque thickness were investigated using univariate and multivariate analyses. The Multivariate analysis included the relevant Univariate results. Crouse score and carotid plaque thickness were substantially correlated with β2-MG, HGF, and Lp-PLA2 levels, as shown in Table 2.

The β2-MG, HGF, Lp-PLA2 levels and outcome

Table 3 summarizes our investigation into the clinical variables linked with this result. The correlation between β2-MG, HGF, and Lp-PLA2 levels and the result was also examined. Correlation was found between high β2-MG levels and the poor prognosis (HR[95%CI], 1.29 [1.03-1.52], $P = .022$). Those patients who had elevated levels of HGF were not likely to do well (hazard ratio [95% CI], 1.38 [1.26-1.56], $P = .015$). In addition, high Lp-PLA2 levels were associated with a poorer prognosis than low levels (HR[95%CI], 1.74 [1.29-2.32], $P = .015$).

DISCUSSION

Cerebral infarction is a common and frequently occurring disease in clinical, and with the intensification of aging, the number of patients is increasing.[9,10] Studies have shown that atherosclerosis, vascular lumen stenosis, spasm,

Figure 1. The association between the β2-MG level and A) Crouse score, B) carotid plaque thickness.

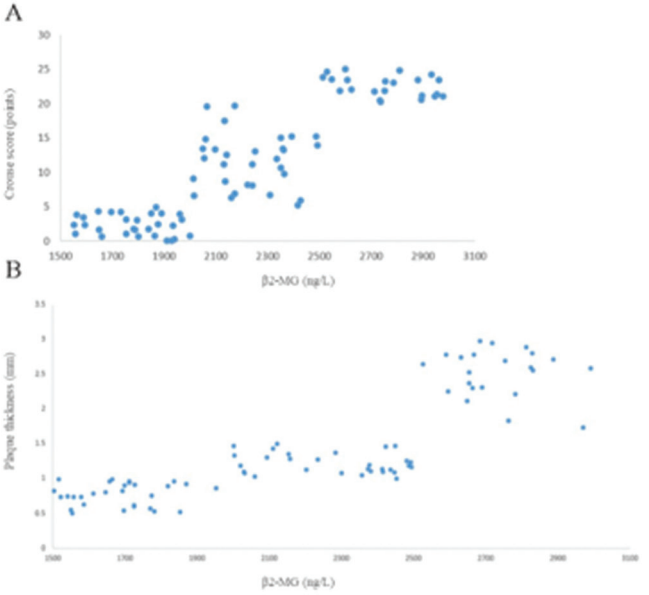

Figure 2. The association between the HGF level and A) Crouse score, B) carotid plaque thickness.

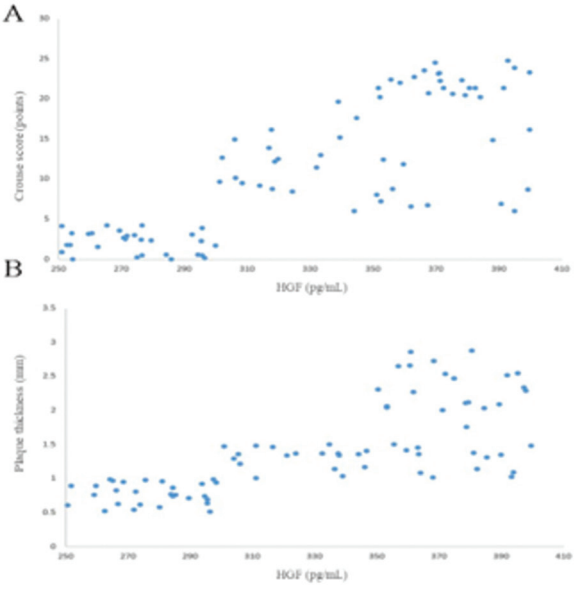

Table 2. The multivariable analyses using the Cox proportional hazard model on carotid plaque-associated factors.

	Crouse score > 15		Carotid plaque thickness > 2mm	
Parameters	HR (95% CI)	*P* value	HR (95% CI)	*P* value
Male sex	1.26 (1.12-1.35)	.022	1.25 (0.88-1.42)	.059
Age	1.52 (1.12-1.78)	.014	1.48 (1.23-1.71)	.012
Diabetes	1.17 (1.09-1.28)	.023	-	-
Platelet	1.33 (1.22-1.48)	.021	1.78 (1.23-2.21)	.027
C-reactive protein	1.54 (1.33-1.68)	.012	1.33 (1.03-1.56)	.034
Blood urea nitrogen	-	-	1.45 (1.19-1.78)	.029
β2-MG	1.59 (1.33-1.81)	.014	1.88 (1.26-2.57)	.017
HGF	1.89 (1.12-2.67)	.078	1.23 (1.12-1.36)	.024
Lp-PLA2	1.77 (1.56-2.12)	.018	1.35 (1.18-1.51)	.018

Figure 3. The association between the Lp-PLA2 level and A) Crouse score, B) carotid plaque thickness.

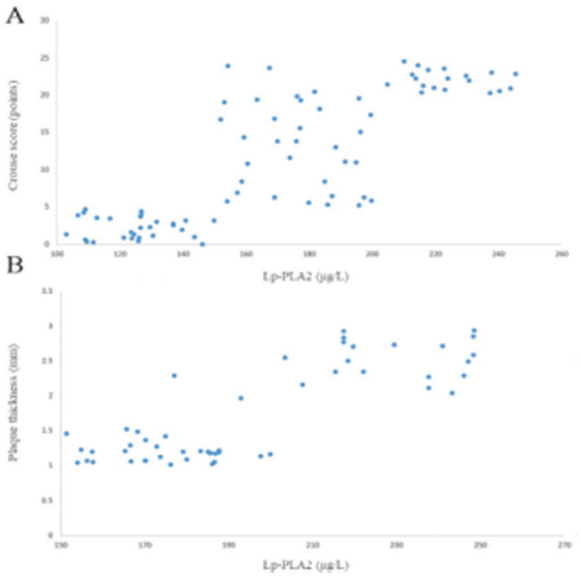

Table 3. The Cox proportional hazard model was used to conduct univariate and multivariate analyses on outcome-related factors.

Factors	Univariate Analysis		Multivariable Analysis	
	HR (95% CI)	*P* value	HR (95% CI)	*P* value
Male (sex)	0.87 (0.65-1.18)	0.387		
Age	0.96 (0.88-1.05)	0.961		
Drinking	1.09 (0.55-1.56)	0.556		
Smoker	0.76 (0.46-1.13)	0.914		
Diabetes	0.60 (0.30-1.22)	0.778		
Chronic heart disease	0.73 (0.16-1.15)	0.256		
ALT	1.11 (0.88-1.38)	0.189		
AST	1.24 (1.08-1.44)	0.578		
Albumin	1.18 (0.79-1.45)	0.448		
Total bilirubin	1.02 (0.98-1.06)	0.163		
Platelet	1.34 (0.88-1.57)	0.542		
Triglycerides	1.12 (0.92-1.32)	0.551		
Cholesterol	1.11 (0.89-1.33)	0.462		
HDL-C	0.75 (0.46-1.22)	0.233		
LDL-C	1.06 (0.45-2.50)	0.296		
FBS	1.40 (0.59-3.31)	0.374		
Hemoglobin A_{1c}	0.85 (0.31-2.31)	0.288		
Creatinine	1.21 (0.84-1.56)	0.087		
C-reactive protein	0.78 (0.55-0.98)	0.041	0.67 (0.45-0.88)	.037
Blood urea nitrogen	0.32 (0.12-0.56)	0.011	0.38 (0.22-0.53)	.008
β2-MG	1.22 (1.09-1.33)	0.032	1.29 (1.03-1.52)	.022
HGF	1.44 (1.26-1.55)	0.017	1.38 (1.26-1.56)	.015
Lp-PLA2	1.88 (1.32-2.19)	0.004	1.74 (1.29-2.32)	.006

and increased blood aggregation and viscosity are the main pathological basis of cerebral infarction, leading to the insufficient blood supply to brain tissue, resulting in brain tissue ischemia and hypoxic necrosis. Cerebral infarction has been presented as the second leading cause of death and the leading cause of disability around the world.[11] Progressive cerebral infarction accounts for 26%-43% of ischemic strokes with a higher disability rate and worse prognosis. The clinical diagnosis of progressive cerebral infarction depends on the change in the degree of neurological deficit in patients from mild to severe. The treatments of cerebral infarction are thrombolysis, anticoagulation, and improving blood viscosity and blood lipid metabolism to reduce brain cell damage, improve brain blood supply, and restore nerve function. There have been many clinical reports on the prognostic factors of cerebral infarction, but the results are controversial and need to be further clarified.[12]

As a growth factor, HGF contributes to developing, repairing, and remodeling many bodily systems. and studies have found that it is also distributed in the nervous system. HGF is involved in the occurrence and development of hypertension and the reconstruction of its formed tissues. It has some local regulatory systems in vascular endothelial and smooth muscle cells. It has played a role in its development, playing an important role in endothelial repair after vascular wall injury and in the gradual formation of neointima.[13-15] Several recent investigations have shown HGF's presence in brain nerve tissue, suggesting this growth factor likely contributes to developing new neuronal substances and coordinating existing neural networks.

β2-MG is a small molecular globulin that was first isolated from the urine of patients with renal tubulopathy by the Swedish chemist BERGGARD in 1968 and excreted by the kidneys. It is the same substance and an important cell membrane surface part. Serum β2-MG is mainly secreted by nucleated cells and lymphocytes.[16] When the metabolism of human histocompatibility antigens is accelerated, or a large number of cells are necrotic, β2-MG is released into the blood from free form, and most of it is passed through the glomerulus.[17,18] Pinocytosis reabsorption decomposes it into amino acids, so the serum β2-MG will increase only when the body is in a pathological state.

LAA and SAO account for most cerebral infarction, and both belong to cerebrovascular disease. Although some risk factors are similar to the existing treatment methods, there are still differences in clinical manifestations and pathology. Lp-PLA2 is an important participant in arterial inflammatory response.[19] The literature has reported that it is related to heart disease, cerebrovascular disease, diabetic nephropathy, asthma, etc. The cytokines mediated by it can promote matrix metalloproteinases in carotid atherosclerotic plaques. The expression of this enzyme increases plaque vulnerability. It can degrade components such as the fibrous cap and collagen matrix in the plaque, leading to plaque rupture and increased internal bleeding. This vulnerability increases the occurrence of ischemic vascular disease. This study's correlation analysis established a link between Lp-PLA2 and atherosclerotic plaques by showing a positive connection between Lp-PLA2 and the crouse score.

In conclusion, the levels of β2-MG, HGF, and Lp-PLA2 in patients with cerebral infarction were substantially linked with carotid plaques.

DATA AVAILABILITY

When requested, the corresponding author will provide access to the simulation experiment data used to support the results of this work.

CONFLICTS OF INTEREST

The authors confirm that they have no financial or personal stake in preventing this research from being published.

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There are no funding sources to declare.

REFERENCE

1. Ikenouchi H, Yoshimoto T, Ihara M. Postprandial cerebral infarction. [J]. *J Clin Neurosci*. 2021;94(11):38-40. doi:10.1016/j.jocn.2021.09.034
2. Sun W, Li G, Zeng X, et al. Clinical and Imaging Characteristics of Cerebral Infarction in Patients with Nonvalvular Atrial Fibrillation Combined with Cerebral Artery Stenosis. [J]. *J Atheroscler Thromb*. 2018;25(8):720-732. doi:10.5551/jat.43240
3. Jin X, Zou Y, Zhai J, Liu J, Huang B. Refractory Mycoplasma pneumoniae pneumonia with concomitant acute cerebral infarction in a child: A case report and literature review. [J]. *Medicine (Baltimore)*. 2018;97(13):e0103. doi:10.1097/MD.0000000000010103
4. Tian Y, Su G, Liu JF, et al. Rodent models for intravascular ischemic cerebral infarction: a review of influencing factors and method optimization. [J]. *Neuroreport*. 2020;31(16):1154-1160. doi:10.1097/WNR.0000000000001529
5. Jiang Q, Xiao S, Shu L, Huang X, Chen X, Hong H. Pituitary Apoplexy Leading to Cerebral Infarction: A Systematic Review. [J]. *Eur Neurol*. 2020;83(2):121-130. doi:10.1159/000507190
6. Nishijo M, Morikawa Y, Nakagawa H, et al. Causes of death and renal tubular dysfunction in residents exposed to cadmium in the environment. [J]. *Occup Environ Med*. 2006;63(8):545-550. doi:10.1136/oem.2006.026591
7. Yamashita T, Abe K. Recent Progress in Therapeutic Strategies for Ischemic Stroke. [J]. *Cell Transplant*. 2016;25(5):893-898. doi:10.3727/096368916X690548
8. Yan P, Cao J, Zhou Y, Zhou X, Sun Z, Zhu X. Serum levels of sLOX-1 and Lp-PLA2 can predict the prognosis of acute cerebral infarction with a high specificity. [J]. *Physiol Rep*. 2022;10(1):e15160. doi:10.14814/phy2.15160
9. Edwards MD, Hughes TAT. Managing blood pressure in acute cerebral infarction. [J]. *J Neurol*. 2021;268(6):2294-2296. doi:10.1007/s00415-021-10622-6
10. Saver JL. Proposal for a universal definition of cerebral infarction. [J]. *Stroke*. 2008;39(11):3110-3115. doi:10.1161/STROKEAHA.108.518415
11. Ueno T, Nishijima H, Hikichi H, et al. Helicopter Transport for Patients with Cerebral Infarction in Rural Japan. [J]. *J Stroke Cerebrovasc Dis*. 2019;28(9):2525-2529. doi:10.1016/j.jstrokecerebrovasdis.2019.06.010
12. Mirvis SE, Wolf AL, Numaguchi Y, Corradino G, Joslyn JN. Posttraumatic cerebral infarction diagnosed by CT: prevalence, origin, and outcome. [J]. *AJR Am J Roentgenol*. 1990;154(6):1293-1298. doi:10.2214/ajr.154.6.2110744
13. Naess H, Romi F. Comparing patients with spinal cord infarction and cerebral infarction: clinical characteristics, and short-term outcome. [J]. *Vasc Health Risk Manag*. 2011;7:497-502. doi:10.2147/VHRM.S22950
14. Büsing KA, Schulte-Sasse C, Flüchter S, et al. Cerebral infarction: incidence and risk factors after diagnostic and interventional cardiac catheterization--prospective evaluation at diffusion-weighted MR imaging. [J]. *Radiology*. 2005;235(1):177-183. doi:10.1148/radiol.2351040117
15. Rizvi F, Everton E, Smith AR, et al. Murine liver repair via transient activation of regenerative pathways in hepatocytes using lipid nanoparticle-complexed nucleoside-modified mRNA. [J]. *Nat Commun*. 2021;12(1):613. doi:10.1038/s41467-021-20903-3
16. Ren L, Zhang C, Wen W, et al. Research on the Application Value of Serum and Urine β2-Microglobulin in Immunoglobulin a Nephropathy. [J]. *Clin Lab*. 2020;66(7):11-17. doi:10.7754/Clin.Lab.2019.191117
17. Wang Q, Zhang M, Zhai M, Li Z. Cerebral infarction caused by systemic sclerosis: a case report. [J]. *J Int Med Res*. 2021;49(9):3000605211044045. doi:10.1177/03000605211044045
18. Lin J, Xian W, Lai R, et al. Bilateral cerebral infarction associated with severe arteriosclerosis in the A1 segment: a case report. [J]. *J Int Med Res*. 2019;47(3):1373-1377. doi:10.1177/0300060519828930
19. Epps KC, Wilensky RL. Lp-PLA_2- a novel risk factor for high-risk coronary and carotid artery disease. [J]. *J Intern Med*. 2011;269(1):94-106. doi:10.1111/j.1365-2796.2010.02297.x