Neutrophil to lymphocyte ratio predicts island sign in patients with intracranial hemorrhage

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Abstract
Our previously studies indicated that inflammatory responses are involved in the hematoma expansion (HE) after intracranial hemorrhage (ICH) ictus. Here, we aim to evaluate the correlations among the ratio of neutrophil to lymphocyte ratio (NLR), HE, and island sign in patients with ICH.

Patients with spontaneous ICH were retrospectively included. Clinical characteristics, imaging features, and laboratory parameters were obtained. Multivariable analysis was performed to evaluate the association of NLR with HE or island sign. Receiver-operator analysis was also used to estimate their predictive abilities for HE and its imaging features.

A total of 279 patients were enrolled in present study, and 78 patients had early hematoma growth, while 43 of them exhibited island sign. Elevation of both leukocyte (odds ratio [OR] 1.136, 95% confidence interval [CI] 1.037–1.245, P < .01) and neutrophil absolute numbers (OR 1.169, 95% CI 1.065–1.284, P < .01), as well as reduction of lymphocyte counts (OR 0.052, 95% CI 0.016–0.167, P < .01) were strongly associated with the existence of island sign. Moreover, despite the predictive ability of NLR on the existence of island sign (OR 1.063, 95% CI 1.036–1.090, P < .01), it also showed the best predictive accuracy (sensitivity 76.74%, specificity 79.66%, positive predictive value 40.70%, negative predictive value 94.90%, area under the curve 0.817) by comparing with peripheral leukocyte counts.

The NLR could be used as an independently marker for reflecting the island sign in patients with ICH. Our findings indicated that systemic inflammatory responses might be involved in the pathologic process of active bleeding in cerebral.

Abbreviations: ALC = absolute lymphocyte count, AMC = absolute monocyte count, ANC = absolute neutrophil count, APTT = activated partial thromboplastin time, AUC = area under the curve, CI = confidence interval, CT = computed tomography, HE = hematoma expansion, ICH = intracranial hemorrhage, INR = international normalized ratio, IQR = interquartile range, IVH = intraventricular hemorrhage, NLR = neutrophil to lymphocyte ratio, OR = odds ratio, PT = prothrombin time, ROC = receiver operating curve, SAH = subarachnoid hemorrhage, sICH = spontaneous intracranial hemorrhage, WBC = white blood count.

Keywords: hematoma expansion, inflammation, intracranial hemorrhage, island sign, neutrophil to lymphocyte

1. Introduction
Spontaneous intracranial hemorrhage (ICH) is one of the most severe diseases thus lead to high ratio of mortality and morbidity in patients. It is well-documented that inflammatory responses and early hematoma growth play important roles in the pathologic processes of ICH. However, the relationship between inflammation and hematoma expansion (HE) remains controversial due to the inconsistent results that observed in previous studies. Recently, neutrophil to lymphocyte ratio (NLR) is employed as a novel inflammatory marker to independently predict the prognosis of patients with ICH. By comparing with the absolute counts of both neutrophil count (ANC) and lymphocyte count (ALC), NLR exhibited the best predictive value for functional outcome in patients with ICH. Here, we aimed to evaluate the potential association of NLR with HE, as well as island sign, which was recently identified as an excellent imaging predictor for HE in patients with ICH.

2. Methods
2.1. Patient selection
All the cases of patients with ICH that admitted in West China Hospital between September 2014 and October 2016 were retrospectively reviewed. Inclusion criteria were as follow: patients were diagnosed as ICH by computed tomography (CT); laboratory tests were performed within 24 hours after admission; initial CT scans were performed within 6 hours after admission and followed up CT scans were performed within 24 hours after initial CT; the age of all patients were older than
evaluate association of NLR and HE as well as island sign. Receiver-operator analysis was applied to estimate the predictive ability of NLR for HE and island sign. The variables were considered statistically significant if \( P < .05 \). All the above-mentioned analyses were performed using SPSS 23.0.

3. Results

A total of 279 cases of patients with ICH, including 207 males and 72 females were enrolled in currently study (Fig. 1). The average age was 56.59 ± 11.95 years ranging from 31 to 89 years. The interval time from symptom onset to perform the initial CT scan was 3.80 ± 2.05 hours. The baseline of hematoma volume was 28.13 ± 16.71 mL. The total number of patients with HE is 78, whereas 75 of them were caused by supratentorial hemorrhage over 259 patients (29.0%), and 3 of patients with HE (15.0%) were due to infratentorial hemorrhage \( (P = .18) \). All the detail information and baseline features of included patients with or without HE are shown in Table 1. In addition, island sign was found in 43 patients (15.4%). The \( \kappa \) value for interobserver reliability of island sign was 93.5%, which suggested an excellent interobserver agreement between 2 neuroradiologists. By comparing with patients with ICH without HE, more island sign could be observed in patients with HE (44.9%). The association between clinical characteristics and island sign was also analyzed (Table 2).

According to the blood tests and further analysis, higher WBC (with HE: without HE = 12.94 ± 3.59: 11.56 ± 4.18, \( P = .011 \)), ANC (with HE: without HE = 10.88 ± 3.52: 9.49 ± 4.09, \( P = .009 \)), as well as NLR (with HE: without HE = 18.78 ± 13.47: 11.28 ± 10.87, \( P < .001 \)) showed in patients with HE (Table 1). Meanwhile, patients that possessed island sign had higher WBC (with island sign: without island sign = 13.66 ± 4.32: 11.63 ± 3.95, \( P = .002 \)), ANC (with island sign: without island sign = 11.94 ± 4.22: 9.51 ± 3.83, \( P < .001 \)), and NLR (with island sign: without island sign = 22.63 ± 11.64: 11.69 ± 11.43, \( P < .001 \)), whereas lower ALC (with island sign: without island sign = 0.65 ± 0.31: 1.21 ± 0.90, \( P < .001 \)) were also observed in patients with HE (Table 2). Based on the results from univariate analysis, we found that WBC, ANC and NLR was positively associated with HE and island sign, respectively (Tables 3 and 4). Furthermore, ALC but not AMC was strongly associated with HE \( (P < .001) \). All the above-mentioned analysis demonstrated that WBC (odds ratio [OR] 1.136, 95% confidence interval [CI] 1.037–1.245, \( P < .01 \)), ANC (OR 1.169, 95% CI 1.065–1.284, \( P < .01 \)) and NLR (OR 1.063, 95% CI 1.036–1.090, \( P < .001 \)) could be used for predicting island sign but not HE (Tables 3 and 4).

Simultaneously, our results also indicated that it is interesting that ALC (OR 0.052, 95% CI 0.016–0.167, \( P < .01 \)) were negatively associated with island sign (Table 4). Following the receiver operating characteristic analysis (ROC) was employed to compare the predictive abilities of relevant inflammatory predictors for island sign in patients with HE. The sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and area under the curve (AUC) of NLR for predicting island sign were 76.74%, 79.66%, 40.70%, 94.90%, and 0.817, respectively, the best predictive cut-off value was 14.53 (Fig. 2). The ROC curves and the areas under the curves (AUCs) of laboratory parameters for predicting island sign are also determined in our study, and results elucidated that NLR harbored the best predictive ability for island sign by comparing with other laboratory values (Fig. 2).
4. Discussion

Based on the results, we concluded that elevated NLR could independently predict island sign but not HE. Moreover, increasing of both WBC and ANC are associated with the existence of island sign, while lower ANC could independently predict the existence of island sign. Over all the predicative parameters, NLR showed the best predictive ability for island sign. Taken together, we systematically demonstrated that NLR was associated with the outcome of patients with ICH with HE, and might be used as a valuable predictive marker for HE and the existence of island sign.

Accumulating evidences indicated that HE triggered inflammation accelerates brain injury after ICH in patients.\[^{8,9,13,14}\] Although the relationship between leucocyte and prognosis in patients with ICH was well documented,\[^{15,16}\] but the link between early HE and subsets of leucocytes was still controver-

sial\[^{9,8,17,18}\]. In our study, after strictly excluded the confounders, we could not find that WBC, ANC, and NLR act as biomarker in early HE, except an elevation of them were observed. However, WBC, ANC, and ALC were detected to have the strong predictive ability in island sign, which represented an efficient neuroimaging predictor for early hematoma enlargement. As one of the latest neuroradiologic features, island sign may reflect the active bleeding and is supposed to depict multifocal small bleeding hematomas around the main hematoma.\[^{12}\] Due to this indirect evidence, we expect that all those biomarkers mentioned above exist potential predicting values of early HE in patients with ICH. However, the exact underlying mechanisms of the links between blood routine variables and HE remain elusive and need to further investigate.

The NLR represents a combined index that reflects systemic immune status. As a novel inflammatory marker, it was recently identified to independently predict the progression and
### Table 1
Baseline clinical characteristics related to hematoma expansion in patients with ICH.

| Characteristic                  | Baseline (n = 270) | Patients with HE (n = 78) | Patients without HE (n = 201) | P     |
|---------------------------------|--------------------|--------------------------|------------------------------|-------|
| Male                            | 207 (74.2)         | 57 (73.1)                | 150 (74.6)                   | .791  |
| Age, y                          | 56.59 ± 11.96      | 57.13 ± 12.27            | 56.39 ± 11.85                | .643  |
| Systolic blood pressure, mm Hg  | 168.61 ± 32.84     | 164.27 ± 27.11           | 170.29 ± 34.73               | .169  |
| Diastolic blood pressure, mm Hg | 97.83 ± 19.82      | 95.55 ± 19.06            | 98.71 ± 20.08                | .233  |
| Mean arterial pressure, mm Hg   | 121.20 ± 22.62     | 117.75 ± 20.16           | 122.54 ± 23.41               | .112  |
| Medical history                 |                    |                          |                              |       |
| Hypertension                    | 165 (59.1)         | 45 (57.7)                | 129 (59.7)                   | .759  |
| Diabetes mellitus               | 21 (7.5)           | 8 (10.3)                 | 13 (6.5)                     | .282  |
| Ischemic stroke                 | 37 (13.3)          | 8 (10.3)                 | 29 (14.4)                    | .357  |
| Smoker                          | 118 (42.3)         | 31 (39.7)                | 87 (43.3)                    | .591  |
| Alcohol (>3 drinks per 24 h)    | 110 (39.4)         | 30 (38.5)                | 80 (39.8)                    | .837  |
| Time to CT, h                   | 3.80 ± 2.05        | 3.14 ± 1.87              | 4.06 ± 2.06                  | .001  |
| GCS score on admission          | 10 (7.13)          | 9 (5.10)                 | 11 (8.14)                    | <.001 |
| Hematoma size, mL               | 28.14 ± 16.71      | 41.49 ± 15.72            | 22.95 ± 14.01                | <.001 |
| Hydrocephalus                   | 50 (17.9)          | 21 (26.9)                | 29 (14.4)                    | .015  |
| Presence of IVH                 | 62 (22.2)          | 24 (30.8)                | 38 (18.9)                    | .032  |
| Supratentorial hematoma          | 259 (92.8)         | 75 (96.2)                | 184 (91.5)                   | .181  |
| Island sign                     | 43 (15.4)          | 35 (44.9)                | 8 (4.0)                      | <.001 |
| PT                              | 10.91 ± 0.65       | 10.94 ± 0.60             | 10.90 ± 0.67                 | .654  |
| APTT                            | 28.13 ± 1.46       | 27.96 ± 1.19             | 28.19 ± 1.54                 | .198  |
| INR                             | 0.92 ± 0.06        | 0.90 ± 0.07              | 0.91 ± 0.06                  | .142  |
| WBC 10^9/L                      | 11.93 ± 4.07       | 12.94 ± 3.59             | 11.56 ± 4.18                 | .011  |
| ANC 10^9/L                      | 9.88 ± 3.08        | 10.88 ± 3.52             | 9.49 ± 4.09                  | .009  |
| ALC 10^9/L                      | 1.12 ± 3.07        | 1.01 ± 0.98              | 1.18 ± 0.72                  | .086  |
| AMC 10^9/L                      | 0.58 ± 0.27        | 0.55 ± 0.25              | 0.58 ± 0.27                  | .265  |
| NLR                             | 13.38 ± 12.11      | 18.78 ± 13.47            | 11.28 ± 10.87                | <.001 |

Data are expressed as n (%), mean ± standard deviation, median (interquartile range), as appropriate.

*ALC = absolute lymphocyte count, AMC = absolute monocyte count, ANC = absolute neutrophil count, APTT = activated partial thromboplastin time, GCS = Glasgow coma scale, HE = hematoma expansion, ICH = intracranial hemorrhage, INR = international normalized ratio, IVH = intraventricular hemorrhage, NLR = neutrophil to lymphocyte ratio, PT = prothrombin time, WBCs = white blood cells.

* Indicates P < .05.

### Table 2
Clinical characteristics related to island sign in patients with ICH.

| Characteristic                  | Patients with island sign (n = 43) | Patients without island sign (n = 236) | P     |
|---------------------------------|-----------------------------------|---------------------------------------|-------|
| Male                            | 32 (74.4)                         | 175 (74.2)                            | .971  |
| Age, y                          | 53.86 ± 12.53                     | 57.03 ± 11.80                         | .103  |
| Systolic blood pressure, mm Hg  | 161.16 ± 28.88                    | 169.97 ± 33.24                        | .106  |
| Diastolic blood pressure, mm Hg | 95.44 ± 17.75                     | 98.26 ± 20.18                         | .392  |
| Mean arterial pressure, mm Hg   | 116.78 ± 19.85                    | 122.01 ± 23.03                        | .164  |
| Medical history                 |                                    |                                       |       |
| Hypertension                    | 23 (53.5)                         | 142 (60.2)                            | .412  |
| Diabetes mellitus               | 7 (16.3)                          | 14 (5.9)                              | .018  |
| Ischemic stroke                 | 4 (9.3)                           | 33 (14.3)                             | .405  |
| Smoker                          | 20 (46.5)                         | 98 (41.5)                             | .543  |
| Alcohol (>3 drinks per 24 h)    | 23 (53.9)                         | 87 (36.9)                             | .041  |
| Time to CT, h                   | 3.31 ± 1.91                       | 3.88 ± 2.06                           | .091  |
| GCS score on admission          |                                    |                                       | .363  |
| Hematoma size, mL               | 36.86 ± 15.64                     | 26.54 ± 16.42                         | <.001 |
| Hydrocephalus                   | 9 (20.9)                          | 41 (17.4)                             | .576  |
| Presence of IVH                 | 9 (20.9)                          | 53 (22.5)                             | .825  |
| PT                              | 10.94 ± 0.58                      | 10.90 ± 0.68                          | .695  |
| APTT                            | 28.58 ± 0.89                      | 28.04 ± 1.52                          | .005  |
| INR                             | 0.91 ± 0.07                       | 0.92 ± 0.06                           | .886  |
| WBC 10^9/L                      | 13.66 ± 4.32                      | 11.63 ± 3.95                          | .002  |
| ANC 10^9/L                      | 11.94 ± 4.22                      | 9.51 ± 3.83                           | <.001 |
| ALC 10^9/L                      | 0.65 ± 0.31                       | 1.21 ± 0.90                           | <.001 |
| AMC 10^9/L                      | 0.64 ± 0.31                       | 0.56 ± 0.25                           | .073  |
| NLR                             | 22.65 ± 11.64                     | 11.69 ± 11.43                         | <.001 |

Data are expressed as n (%), mean ± standard deviation, median (interquartile range), as appropriate.

*ALC = absolute lymphocyte count, AMC = absolute monocyte count, ANC = absolute neutrophil count, APTT = activated partial thromboplastin time, GCS = Glasgow coma scale, ICH = intracranial hemorrhage, INR = international normalized ratio, IVH = intraventricular hemorrhage, NLR = neutrophil to lymphocyte ratio, PT = prothrombin time, WBCs = white blood cells.

* Indicates P < .05.
outcome of many diseases in human patients.\textsuperscript{19–22} Accumulating results from several studies reported that NLR was associated with hematoma size,\textsuperscript{23} 30-day mortality,\textsuperscript{24} 90-day mortality,\textsuperscript{25} and poor outcome\textsuperscript{8} in patients with IHC. However, whether NLR could be also used for predicting HE in patients with ICH remains unknown. Here, we revealed that NLR was associated with island sign and showed the best predictive value of this neuroradiologic feature by comparing with other laboratory parameters. But the irrelevance of NLR with hematoma growth in patients with ICH was also observed. The possible reasons of this phenomenon were as
follow: firstly, elevating NLR could induce neurotoxicity thus activated the matrix metalloproteinases, which triggered the basal membrane components degradation, BBB breakdown, and brain edema and active bleeding. Secondly, inflammatory cascades were activated by thrombin during HE and resulted the elevation of WBC count, C-reactive protein, and interleukin-6, which further promotes HE. At last, cellular immune response was reported to damage coagulation function and microvascular integrity via inducing C-reactive protein (CRP) activation, which is a relatively chronic process.

Therefore, it is expected that systemic increasing protein (CRP) activation, which is a relatively chronic function and microvascular integrity via inducing C-reactive cellular immune response was reported to damage coagulation interleukin-6, which further promotes HE. At last, cellular immune response was reported to damage coagulation function and microvascular integrity via inducing C-reactive protein (CRP) activation, which is a relatively chronic process.

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References

[1] Qureshi AI, Mendelow AD, Hanley DF. Intracerebral haemorrhage. Lancet 2009;373:1632–44.

[2] Qureshi AI, Tuhrim S, Broderick JP, et al. Spontaneous intracerebral hemorrhage. N Engl J Med 2001;344:1450–60.

[3] Aronowski J, Zhao X. Molecular pathophysiology of cerebral hemorrhage: secondary brain injury. Stroke 2011;42:1781–6.

[4] Brouwers HB, Zhang Y, Falcone GJ, et al. Predicting hematoma expansion after primary intracerebral hemorrhage. JAMA Neurol 2014;71:158–64.

[5] Chai S, Correll C, Veerina KT, et al. Prediction of intracerebral haemorrhage expansion with clinical, laboratory, pharmacologic, and noncontrast radiographic variables. Int J Stroke 2015;10:1057–61.

[6] Chen S, Zhao B, Wang W, et al. Predictors of hematoma expansion predictors after intracerebral hemorrhage. Oncotarget 2017;8:89548–63.

[7] Yu Z, Zheng J, Ali H, et al. Significance of satellite sign and spot sign in predicting hematoma expansion in spontaneous intracerebral hemorrhage. Clin Neurol Neurosurg 2017;162:67–71.

[8] Lattanzio S, Gagnetti C, Ponzoni M, et al. Neutrophil-to-lymphocyte ratio predicts the outcome of acute intracerebral hemorrhage. Stroke 2016;47:1654–7.

[9] Silva Y, Leira R, Tejada J, et al. Molecular signatures of vascular injury are associated with early growth of intracerebral hemorrhage. Stroke 2005;36:86–91.

[10] Kothari RU, Brott T, Broderick JP, et al. The ABCs of measuring intracerebral hemorrhage volumes. Stroke 1996;27:1304–5.

[11] Davis SM, Broderick J, Hennerici M, et al. Hematoma growth is a determinant of mortality and poor outcome after intracerebral hemorrhage. Neurology 2006;66:1175–81.

[12] Li Q, Liu QJ, Yang WS, et al. Island sign: an imaging predictor for early hematoma expansion and poor outcome in patients with intracerebral hemorrhage. Stroke 2017;48:3019–25.

[13] Kuhlmann CR, Librizzi L, Closhen D, et al. Mechanisms of C-reactive protein-induced blood-brain barrier disruption. Stroke 2009;40:1458–66.

[14] Lee KR, Colom GP, Retz AL, et al. Edema from intracerebral hemorrhage: the role of thrombin. J Neurosurg 1996;84:91–6.

[15] Leira R, Davalos A, Silva Y, et al. Early neurologic deterioration in intracerebral hemorrhage: predictors and associated factors. Neurology 2004;63:461–7.

[16] Sun W, Peacock A, Becker J, et al. Correlation of leukocytosis with early neurological deterioration following supratentorial intracerebral hemorrhage. J Clin Neurosci 2012;19:1096–100.

[17] Suzuki S, Kelley RE, Dandapani BK, et al. Acute leukocyte and temperature response in hypertensive intracerebral hemorrhage. Stroke 1995;26:1020–3.

[18] Morotti A, Phua CL, Anderson CD, et al. Leukocyte count and intracerebral hemorrhage expansion. Stroke 2016;47:1473–8.

[19] Cruimley AB, McMillan DC, McKernan M, et al. Evaluation of an inflammation-based prognostic score in patients with inoperable gastrointestinal cancer patients. Cancer Med 2014;3:406–15.

[20] Tokgoz S, Keskin S, Kayrak M, et al. Neutrophil-to-lymphocyte ratio predicts to short-term mortality in acute cerebral infarct independently from infarct volume? J Stroke Cerebrovasc Dis 2014;23:2163–8.

[21] Brooks SD, Spears C, Cummings C, et al. Admission neutrophil-to-lymphocyte ratio predicts 90-day outcome after endovascular stroke therapy. J Neurointerv Surg 2014;6:578–83.

[22] Giede-Jeppe A, Bobinger T, Gerner ST, et al. Neutrophil-to-lymphocyte ratio is an independent predictor for in-hospital mortality in spontaneous intracerebral hemorrhage. Cerebrovasc Dis 2017;44:26–34.

[23] Wang F, Hu S, Ding Y, et al. Neutrophil-to-lymphocyte ratio and 30-day mortality in patients with acute intracerebral hemorrhage. J Stroke Cerebrovasc Dis 2016;25:182–7.

[24] Tao C, Wang J, Hu X, et al. Clinical value of neutrophil to lymphocyte and platelet to lymphocyte ratio after aneurysmal subarachnoid hemorrhage. Neurocrit Care 2017;26:393–401.
[26] Nguyen HX, O’Barr TJ, Anderson AJ. Polymorphonuclear leukocytes promote neurotoxicity through release of matrix metalloproteinases, reactive oxygen species, and TNF-alpha. J Neurochem 2007;102:900–12.

[27] Mayer SA, Lignelli A, Fink ME, et al. Perilesional blood flow and edema formation in acute intracerebral hemorrhage: a SPECT study. Stroke 1998;29:1791–8.

[28] Xi G, Wagner KR, Keep RF, et al. Role of blood clot formation on early edema development after experimental intracerebral hemorrhage. Stroke 1998;29:2580–6.

[29] Roberts CJ, Birkenmeier TM, McQuillan JJ, et al. Transforming growth factor beta stimulates the expression of fibronectin and of both subunits of the human fibronectin receptor by cultured human lung fibroblasts. J Biol Chem 1988;263:4586–92.