Acute neuromyelitis optica spectrum disorder patients' clinical analysis of disability-related biomarkers

Xingyue Zheng
  First Affiliated Hospital of Dalian Medical University

Hongjing Yan
  Handan First Hospital

Hao Yin
  Jining NO.1 People's Hospital

Jing Shi
  Jining NO.1 People's Hospital

Yuanyuan Liu
  Jining NO.1 People's Hospital

Haotian Zhao
  Jining NO.1 People's Hospital

Yuzhi Li
  Jining NO.1 People's Hospital

Huakun Liu
  Jining NO.1 People's Hospital

Lei Zhang
  Jining NO.1 People's Hospital

Zhongrui Yan
  Jining NO.1 People's Hospital

Chunbo Dong
  dcb101@sina.com

  First Affiliated Hospital of Dalian Medical University

Additional Declarations: No competing interests reported.

Abstract

Background: The clinical features of neuromyelitis optica spectrum disorder (NMOSD) predominantly include optic neuritis and myelitis, among other symptoms. A greater level of disability during the acute phase typically suggests an unfavorable prognosis. Nevertheless, the clinical biomarkers that impact the severity of disability in NMOSD remain unclear.

Methods: We analyzed 41 NMOSD patients and 41 normal controls to identify biomarkers associated with the disease. NMOSD patients were categorized into two groups based on their Expanded Disability Status Scale(EDSS) score: mild to moderate disability (EDSS <4) and severe disability (EDSS ≥4). Correlation and ROC analyses were conducted on various biomarkers, including neutrophil-to-lymphocyte ratio (NLR), platelet-to-lymphocyte ratio (PLR), monocyte-to-lymphocyte ratio(MLR), cerebrospinal fluid (CSF)/serum albumin quotient(QAlb), CSF/blood immunoglobulin G quotient (QIgG), CSF/blood immunoglobulin A quotient (QIgA), CSF/blood immunoglobulin M quotient (QIgM), to identify markers linked to disability severity and confirm their independence.

Results: 1. Significant differences in blood NLR, PLR, and MLR were found between NMOSD patients and normal controls (P<0.01) in biomarker comparison analysis. 2. Significant variations in QAlb, QIgG, QIgA, QIgM, and PLR were noted between the two groups of NMOSD patients stratified by disability severity. 3. A correlation analysis revealed a positive association between QAlb, QIgG, QIgA, QIgM, PLR, and EDSS scores. 4. Levels of QAlb, QIgG, QIgA, QIgM, and PLR were found to be effective indicators of NMOSD severity in Receiver Operating Characteristic (ROC) analysis (P<0.01). 5. Multifactor regression analysis confirmed the independence of PLR in assessing disease severity (P<0.01).

Conclusion: 1. QAlb, QIgG, QIgA, QIgM, and PLR have demonstrated efficacy as biomarkers for assessing the severity of NMOSD; 2.PLR has shown promise as a standalone indicator for evaluating disease severity in patients with NMOSD.

1 Introduction

NMOSD is a rare yet severe neuroinflammatory disorder characterized by inflammation primarily impacting the optic nerves and spinal cord, resulting in significant consequences such as vision loss, as well as motor and sensory impairments [1]. The pathogenesis of NMOSD is believed to involve the binding of immunoglobulin G anti-aquaporin-4 autoantibodies (NMO-IgG) to astrocytes, which induces complement-dependent cytotoxicity (CDC) and subsequent inflammation, demyelination, and neuronal loss[2]. However, studies have indicated that there isn't necessarily a positive correlation between serum AQP4 antibody levels and the severity of acute disability in patients. This inconsistency may be attributed to variations in the degree of blood-brain barrier (BBB) disruption among different patients.

BBB is a critical component of the neuroimmune axis. There is a strong correlation between the severity of immune-related central nervous system (CNS) disorders and BBB disruption. BBB disruption impedes the controlled exchange of chemokines, cytokines, and immune cells between the CNS and the bloodstream, leading to increased disorganized exchange of substances such as IgG and plasma albumin across the brain-blood interface. BBB disruption is a prominent pathological feature observed in individuals with NMOSD. Animal experiments have also confirmed the significance of BBB disruption in the occurrence and progression of NMOSD [3, 4]. Numerous clinical studies have demonstrated a strong correlation between the extent of BBB disruption and the severity of disability in NMOSD. This can be evaluated by measuring the QAlb to gauge the extent of BBB damage. Research outcomes suggest that a higher QAlb ratio is linked with heightened disability severity in NMOSD patients[5, 6, 7] .

Additionally, previous studies have indicated that neutrophils, platelets, and monocytes in the blood serve as important biomarkers of systemic inflammation. Neutrophils play a role in the initiation stage of inflammatory responses[8]. Platelets form platelet-leukocyte aggregates in inflamed tissues by secreting adhesive factors, further promoting and triggering inflammation. Yan's study found that platelets secrete immune regulatory factors which exert chemotactic effects on neutrophils and lymphocytes[9]. Furthermore, studies have indicated that specific blood biomarkers associated with lymphocytes, such as PLR, NLR, and MLR, can serve as indicators of inflammation linked to demyelinating disorders of the central nervous system and other autoimmune conditions[2, 10, 11]. Song JE et al further corroborated this finding, highlighting PLR as a valuable indicator of both inflammatory status and immune activity in patients[12]. Moreover, in Miguel Cabanillas-Lazo's meta-analysis, proposed an additional perspective, suggesting that NLR could also serve as a predictive factor for the prognosis of NMOSD[13]. The relationship between blood inflammatory markers and the severity of NMOSD disability remains unclear. Therefore, it is essential to explore the correlation

between changes in plasma inflammatory biomarker levels, variations in blood-brain barrier disruption indicators, and the severity of disability in NMOSD. A thorough examination of the interactions between these biochemical markers and NMOSD could provide healthcare professionals with improved diagnostic tools and innovative approaches for the treatment and management of individuals with NMOSD.

This study aims to explore biomarkers associated with the severity of disability in NMOSD and investigate the relationship between these biomarkers and the level of disability in NMOSD patients.

2 Methods

2.1 Study population

A retrospective analysis was conducted on cases from November 2020 to March 2024, including patients with NMOSD. Diagnosis of NMOSD followed international consensus standards[1]. Exclusion criteria were: (a) Patients with comorbidities such as hypertension, coronary heart disease, diabetes, cerebrovascular disease, tumors, and other chronic diseases; (b) Patients with other autoimmune diseases such as rheumatoid arthritis, Sjogren's syndrome, inflammatory myositis, systemic vasculitis, systemic lupus erythematosus, and thyroid diseases.

2.2 Study Design

This is a single-center retrospective observational cohort study. We collected demographic and clinical data, including variables such as age, gender, AQP4-IgG status, clinical symptoms, medication usage, history of attacks, EDSS scores, QAlb, QIgG, QIgA, QIgM, and cerebrospinal fluid immunoglobulin G index. QAlb was utilized as an indicator of blood-brain barrier permeability in the study. the formula used to calculate QAlb is QAlb = CSF albumin/serum albumin$\times 10^{-3}$. Considering that QAlb is affected by age, the upper limit of QAlb was adjusted using the formula 4 + (age/15 years)[14]. If QAlb exceeds the normal range, it is classified as increased blood-brain barrier permeability, whereas if QAlb falls within the normal range, it is classified as normal. Cerebrospinal fluid albumin was analyzed using isoelectric focusing, and serum albumin was determined using the latex-enhanced turbidity method. Cerebrospinal fluid and blood samples were collected at admission and before the commencement of any therapeutic interventions. NLR, PLR, and MLR were calculated using peripheral blood cell counts. The neurological impairment of patients upon admission was evaluated by two researchers utilizing the EDSS score[15]. Previous research has indicated that patients can be categorized into two groups based on their EDSS scores: 0-3.5 representing mild to moderate disability, and 4-9.5 indicating severe disability[16, 17]. To explore the independent association between risk factors and disease severity, patients were divided into two cohorts: the mild to moderate NMOSD group (EDSS < 4, N = 23) and the severe NMOSD group (EDSS ≥ 4, N = 18). Typically, patients are considered to be in the acute phase of their illness if the disease duration is less than 30 days. ROC curves were used to assess the diagnostic accuracy of the test based on EDSS scores, with the optimal cutoff value determined using the Youden Index. Logistic regression analysis was employed for both univariate and multivariate analyses to evaluate the independent factors influencing NMOSD disability severity.

2.3 Standard protocol approval, registrations, and patient consent

This study received approval from the Medical Ethics Committee of Jining No.1 People's Hospital (KYLL-202307-102), and written informed consent, including neurological consent, was obtained from all participants or their legal guardians. The research adhered to the principles outlined in the Helsinki Declaration and was conducted by approved guidelines.

2.4. Statistical Analysis

The research data were analyzed using SPSS 22.0 software, where the Kolmogorov-Smirnov test revealed a non-normal distribution. Data visualization was performed using GraphPad Prism. Spearman's non-parametric correlation test was chosen to evaluate the relationship between continuous variables, given the non-normal distribution of the data. Normally distributed variables were expressed as mean ± standard deviation (SD) and compared using Student's t-test. Non-normally distributed data were represented by the median and interquartile range (25−75%) and analyzed using the Mann-Whitney U-test. Categorical variables were depicted as percentages (%) and compared between groups using the chi-square test. Receiver Operating Characteristic (ROC) curves were utilized to establish critical values. Both univariate and multivariate logistic regression models were applied, with a significance level set at P < 0.05 considered statistically significant.

2.5 Data availability

The datasets used and analyzed during the current study are available from the corresponding author upon reasonable request.

3 Results

3.1 Inter-group Clinical Characteristics Analysis

Table 1 presents a comparison of clinical measurements between 41 NMOSD patients (16 males and 25 females) and 41 healthy control subjects (14 males and 27 females) included in the study. While no significant disparities were found in age and gender between the two groups, variations were noted in the levels of NLR, PLR, and MLR. Figure 1 illustrates the comparison of blood inflammatory markers between NMOSD patients and the normal control group. The results indicate that NLR (P = 0.01), PLR (P < 0.001), and MLR (P < 0.001) levels in NMOSD patients were significantly higher than those in the healthy control group.

Table 1
The demographics and clinical characteristics of participants between NMOSD group and the healthy control group

Clinical characteristics	Healthy control group		NMOSD group		*P*-values
	N = 41		N = 41		
Sex,(N,%)	male	female	male	female	0.819
	14(34.15%)	27(65.85%)	16(39.02%)	25(50.98%)	
Age, y (mean ± SD)	41.76 ± 8.932		40.88 ± 15.330		0.751
AQP4-IgG + of patients(n,%)	—		15(36.58%)		—
Optic neuritis(n,%)	—		6(14.63%)		—
Acute myelitis(n,%)	—		29(70.74%)		—
Mix attacks(n,%)	—		6(14.63%)		—
EDSS<4(n,%)	—		21(51.21%)		—
EDSS ≥ 4(n,%)	—		20(48.79%)		—
NLR(M, IQR)	1.65 (1.06, 2.05)		1.89 (1.52, 3.60)		0.01
MLR(M, IQR)	0.12 (0.08, 0.19)		0.88 (0.21, 1.57)		<0.001
PLR(M, IQR)	76.96 (65.92, 98.56)		127.22 (100.82, 163.30)		<0.001

NMOSD, neuromyelitis optica spectrum disorder; SD, standard deviation; *Mix attacks*, including equal to two or more two damaged sites; *AQP4*,aquaporin-4; *EDSS*, Expanded Disability Status Scale; *NLR*,neutrophil-to-lymphocyte ratio; *MLR*, lymphocyte to monocyte ratio; *PLR*, platelet count to lymphocyte ratio;

3.2 Clinical Characteristics Analysis Based on EDSS Score Groups

In this study, patients were divided into two cohorts according to their EDSS scores: EDSS ≥ 4 (n = 20) and EDSS < 4 (n = 21). The male-to-female ratio was similar between the two groups. There were no statistically significant differences observed in mean age, AQP4-IgG positivity, specific symptoms, cerebrospinal fluid IgG index, NLR value, and MLR value between the two groups. However, significant differences were observed between the EDSS < 4 group and the EDSS ≥ 4 group in PLR value (P < 0.001), QAlb value (P = 0.005), QIgG value (P = 0.002), QIgA value (P = 0.016), and QIgM value (P = 0.002) (Table 2, Fig. 2).

Table 2
Demographic and clinical differences between EDSS < 4 and EDSS ≥ 4 in patients with acute NMOSD

Clinical characteristics	EDSS<4		EDSS ≥ 4		P-value
	N = 21		N = 20		
Sex,(N,%)	male	female	male	female	> 0.999
	13(61.90%)	8(38.10%)	12(60.00%)	8(40.00%)	
Age,y (mean ± SD)	33.33 ± 672		37.44 ± 15.545		0.453
AQP4-IgG + of patients(n,%)	7(33.33%)		8(40%)		0.906
Focus:					0.172
Optic neuritis(n,%)	4(19.04%)		2(10.00%)		
Acute myelitis(n,%)	16(76.19%)		13(65.00%)		
Mix attacks(n,%)	1(4.77%)		5(25.00%)		
QAlb (M, IQR)	4.74 (4.04, 5.24)		7.17 (5.20, 9.68)		0.005
QIgG (M, IQR)	2.43 (1.95, 2.74)		3.97 (2.68, 5.50)		0.002
QIgA (M, IQR)	1.41 (1.07, 1.60)		1.99 (1.29, 3.23)		0.016
QIgM (M, IQR)	0.28 (0.18, 0.50)		0.78 (0.49, 1.12)		0.002
CSFIgG index (M, IQR)	0.49 (0.41, 0.54)		0.55 (0.50, 0.60)		0.072
NLR(M, IQR)	1.71 (1.23, 2.86)		2.25 (1.59, 3.84)		0.190
MLR(M, IQR)	1.19 (0.26, 1.54)		0.29 (0.21, 1.41)		0.306
PLR(M, IQR)	101.02 (94.21, 116.45)		157.93 (133.70, 230.28)		< 0.001

3.3 Relationship between Biomarkers and EDSS Score

Correlation analysis in the study examined the relationship between each biomarker and EDSS score, as depicted in Fig. 3. QAlb (r = 0.4102; P = 0.0077), QIgG (r = 0.5435; P = 0.0002), QIgA (r = 0.4428; P = 0.0037), QIgM (r = 0.5801; P = 0.0001), and PLR (r = 0.5887; P = 0.0001) were all positively correlated with EDSS score. NLR (r=-0.1132; P = 0.4809) and MLR (r = 0.0057; P = 0.9717) showed no significant correlation with the EDSS score.

3.4 Predictive Value of Biomarkers for the Severity of EDSS Scores in NMOSD Patients

Receiver Operating Characteristic (ROC) analysis was conducted to assess the predictive value of each biomarker for the severity of EDSS scores in NMOSD patients. ROC analysis was conducted to determine the optimal cut-off values, sensitivity, and specificity of QAlb, QIgG, QIgA, QIgM, NLR, PLR, and MLR in differentiating between NMOSD patients with EDSS scores ≥ 4 and < 4, to categorize disease severity. As shown in Fig. 4: QAlb (cut-off value: 6.3; AUC: 0.8213; sensitivity: 65%; specificity: 90.48%; 95% confidence interval: 0.6726−0.9699; P < 0.001), QIgG (cut-off value: 2.685; AUC: 0.8116; sensitivity: 83.33%; specificity: 73.91%; 95% confidence interval: 0.66631−0.9601; P < 0.001), QIgA (cut-off value: 2.045; AUC: 0.7585; sensitivity: 55.56%; specificity: 100%; 95% confidence interval: 0.5897−0.9272; P < 0.01), and PLR (cut-off value: 120.2; AUC: 0.8116; sensitivity: 65%; specificity: 90.48%; 95% confidence interval: 0.6786−0.9446; P < 0.001) all demonstrated good specificity and sensitivity, allowing prediction of the severity of EDSS scores in patients. They possess clinical predictive significance. NLR (AUC: 0.5821; P = 0.3718) and MLR (AUC: 0.5676; P = 0.4620) showed no significant correlation with the severity of EDSS scores and had no clinical predictive value.

ROC curve analysis was used to distinguish between acute NMOSD patients with EDSS score ≥ 4 and those with EDSS score < 4 using QAlb, QIgG, QIgA, QIgM, NLR, MLR, and PLR. Optimal cut-off values for QAlb, QIgG, QIgA, QIgM, and PLR levels were determined based on the ROC curve area to predict acute NMOSD with EDSS ≥ 4 accurately. The optimal cut-off values for QAlb,

QIgG, QIgA, and PLR show good specificity and sensitivity in predicting disability severity in patients based on EDSS scores, making them clinically valuable for prediction. NLR and MLR do not have a significant correlation with EDSS scores, showing no clinical predictive value.

3.5 Univariate and Multivariate Regression Analysis of Biomarkers to Validate Independent Factors Influencing Disability Severity

Logistic regression analysis was utilized to perform both univariate and multivariate retrospective analyses on biomarkers potentially influencing NMOSD severity. The results presented in Table 3 indicate that PLR (P = 0.01) can function as an independent indicator of NMOSD severity. Conversely, QAlb, QIgG, QIgA, QIgM, NLR, disease location, and AQP4 positivity did not demonstrate statistically significant associations.

Table 3
Univariate and multivariate analysis

Variables	Univariate					Multivariate				
	β	S.E	Z	*P*	OR (95%CI)	β	S.E	Z	*P*	OR (95%CI)
QAlb	0.49	0.19	2.58	0.005	1.63 (1.12 ~ 2.36)	0.94	1.65	0.57	0.567	2.57 (0.10 ~ 65.15)
QIgG	0.91	0.35	2.6	0.002	2.48 (1.25 ~ 4.91)	-1.27	3.39	-0.37	0.709	0.28 (0.00 ~ 217.33)
QIgA	1.34	0.58	2.32	0.016	3.80 (1.23 ~ 11.77)	0.16	2.64	0.06	0.952	1.17 (0.01 ~ 205.90)
QIgM	3.43	1.28	2.69	0.002	30.85 (2.53 ~ 376.19)	2.84	3.57	0.8	0.426	17.11 (0.02 ~ 18612.63)
CSF-IgG-index	2.62	2.4	1.09	0.072	13.75 (0.12 ~ 1526.51)	12.19	17.19	0.71	0.479	195867.25 (0.00 ~ 84690696059175895040.00)
NLR	-0.01	0.1	-0.07	0.190	0.99 (0.82 ~ 1.20)	-0.18	0.18	-0.99	0.322	0.84 (0.59 ~ 1.19)
PLR	0.04	0.01	2.86	**<0.001**	1.04 (1.01 ~ 1.07)	0.06	0.02	2.58	**0.01**	1.06 (1.01 ~ 1.11)
Lesion location	1.1	0.67	1.65	0.099	3.00 (0.81 ~ 11.09)	-0.95	1.38	-0.69	0.492	0.39 (0.03 ~ 5.81)
AQP4	0.29	0.65	0.44	0.658	1.33 (0.37 ~ 4.77)	0.11	1.27	0.08	0.933	1.11 (0.09 ~ 13.45)

QAlb,albumin quotient; *QIgG*, CSF/serum total IgG ratio; *QIgM*, CSF/serum total IgM ratio; *QIgA*, CSF/serum total IgA ratio; *NLR*, neutrophil-to-lymphocyte ratio; *MLR*, lym-phocyte to monocyte ratio; *PLR*, platelet count to lymphocyte ratio;AQP4,aquaporin-4;

OR, Odds Ratio; CI, Confidence Interval.

4 Discussion

In the acute phase of NMOSD, disability scores play a crucial role in predicting patient outcomes. Exploring factors and biomarkers associated with acute-phase disability severity is vital for identifying treatment targets for patients. This study found that QAlb, QIgG, QIgA, QIgM, and PLR are effective in assessing the degree of disability in NMOSD. It is noteworthy that this study revealed the potential of PLR as an independent indicator for evaluating the severity of NMOSD in patients.

QAlb, as a biomarker of blood-brain barrier disruption, has attracted significant attention. QAlb has been considered the gold standard for assessment of the BBB function[18]. Additionally, studies by Chen and Yuan revealed a positive correlation between serum albumin levels and the severity of NMOSD, further validating the association of QAlb with blood-brain barrier damage[19, 20]. In our study, a subgroup analysis of EDSS scores was conducted, and the results showed no significant differences among groups in demographic and basic clinical characteristics. However, compared to patients with EDSS scores<4, those with EDSS scores≥4 exhibited significantly elevated levels of QAlb, QIgG, QIgA, and QIgM. Correlation analysis revealed a positive relationship between the elevated levels of QAlb, QIgG, QIgA, and QIgM and the severity of NMOSD. Our study findings also confirm that during NMOSD, disruption of the blood-brain barrier (BBB) leads to the entry of proteins from the bloodstream into the cerebrospinal fluid, resulting in increased concentrations of albumin and immunoglobulins in the cerebrospinal fluid, thereby elevating QAlb, QIgG, QIgA, and QIgM values. Therefore, higher values of QAlb, QIgG, QIgA, and QIgM reflect greater BBB disruption and more severe disability in patients. This discovery further underscores a direct association between the severity of the disease's disability and the extent of blood-brain barrier damage.

Based on previous research on blood inflammatory markers, the platelet-lymphocyte ratio (PLR) and neutrophil-lymphocyte ratio (NLR) have been evaluated as potential biomarkers for inflammation, disease activity, and prognosis in autoimmune diseases[21, 22]. Our study found significant differences in inflammatory markers (PLR, NLR, MLR) between patients with NMOSD and healthy individuals, indicating that inflammatory response and immune dysregulation are key factors in the development of NMOSD.

Carnero Contentti E et al found that the interaction between the endocrine system and the immune system may significantly promote the onset of NMOSD, indicating that elevated levels of PLR in the blood are often closely associated with the severity and prognosis of NMOSD[23]. Our study results are consistent with those of Carnero Contentti E's study. In subgroup analysis, patients with EDSS scores≥4 showed significantly elevated levels of the inflammatory marker PLR compared to those with EDSS scores<4 (P<0.001). Correlation analysis demonstrates a positive relationship between PLR and disease severity, indicating a close association between platelet alterations and the severity of NMOSD within the context of inflammatory responses. The underlying mechanism may involve platelets forming platelet-leukocyte aggregates in inflamed tissues through the secretion of adhesive factors, further promoting and triggering inflammation. Studies by Yan et al and Chang et al have identified high expression of intercellular adhesion molecule 2 (ICAM2) on platelets, which plays a role in chemotaxis for neutrophils and lymphocytes, facilitating neutrophil crawling and intercellular extravasation[9, 24]. The binding of neutrophils to endothelial cell ICAM2 increases vascular permeability. This indicates that the mutual influence between platelets and inflammatory cells also plays an important role in the disability level of NMOSD. Additionally, the elevation of platelets may be closely associated with changes in the expression levels of matrix metalloproteinase-9 (MMP-9) and claudin-5. Increased platelets may promote the release and activation of MMP-9 by inducing inflammation and damaging endothelial cells, leading to increased degradation of structural proteins in the vascular basement membrane and increased BBB permeability[25]. Moreover, platelet-induced inflammatory responses may also lead to a decrease in the expression levels of claudin-5, affecting the integrity of tight junctions between brain microvascular endothelial cells and increasing BBB permeability[26, 27]. These combined factors collectively lead to increased blood-brain barrier permeability, exacerbating the sensitivity of NMOSD lesion sites to external factors. This makes harmful substances, inflammatory mediators, and immune cells more likely to enter neural tissues, resulting in neuroinflammation and nerve damage. Both Carnero Contentti E and our study confirm the close correlation between PLR levels and the severity of disability in NMOSD patients[23].

In their meta-analysis, Miguel Cabanillas-Lazo et al proposed that NLR could also serve as a prognostic factor for NMOSD. In our study, although NLR showed significant differences between NMOSD and control populations, further subgroup analysis and single-factor/multi-factor regression analysis did not reveal a significant correlation with EDSS scores. This discrepancy may be attributed to sample size and population distribution variations. The differing clinical outcomes from Miguel Cabanillas-Lazo et al also underscore the complexity of the underlying pathophysiology of NMOSD disability, indicating the need for further clinical analysis and validation[13].

Fang X et al found a significant correlation between PLR and the onset of NMOSD[28]. In our ROC analysis, QAlb, QIgG, QIgA, QIgM, PLR, and other biomarkers demonstrated good clinical predictive value in determining the severity of disability, further confirming the clinical relevance between NMOSD disability and biomarkers. Multivariable logistic regression analysis further confirmed that PLR could serve as an independent predictor of NMOSD severity. Yan et al suggested that QAlb might serve as an independent prognostic indicator for disability in NMOSD patients[29]. However, our study results do not support this conclusion. The

inconsistency in results may stem from differences in sample size, population demographics, and the number of influencing factors in multivariate regression analysis.

Sonia D'Souza et al.'s research indicates that platelets are the first blood elements to enter the brain from the peripheral blood after BBB damage in patients with MS. Early-stage prevention of increased platelets in peripheral blood can prevent disease progression[30]. Both NMOSD and MS stem from immune-mediated inflammatory reactions, sharing certain similarities in their pathogenesis. This study demonstrates that PLR can serve as an independent influencing factor affecting the severity of NMOSD, highlighting the significant impact of platelets on the prognosis of NMOSD disability. Therefore, further research should delve into the effects of medications on peripheral blood platelet count and PLR values, as well as their impact on the severity of NMOSD in patients. Such studies can contribute to laying the foundation for personalized clinical interventions and individualized treatment strategies for NMOSD.

Conclusion

This study identified QAlb, QIgG, QIgA, QIgM, and PLR as effective biomarkers for assessing the severity of NMOSD. In particular, PLR emerged as a potential independent indicator for evaluating the severity of NMOSD in patients. These findings provide new insights and methods for the clinical assessment of NMOSD disability, aiding in a more precise evaluation of patient conditions and prognosis. Moreover, the discovery of these disability-related biomarkers in NMOSD opens up possibilities for providing more effective interventions to reduce the severity of disability and improve outcomes.

Limitation

Despite our efforts to ensure the representativeness of the samples and the accuracy of the data, the relatively small sample size in this study may affect the stability and reliability of the results. Additionally, being a single-center study, there may be limitations related to regional and population-specific factors, as well as potential selection bias.

Abbreviations

NMOSD	Neuromyelitis Optica Spectrum Disorder
EDSS	Expanded Disability Status Scale
NLR	Neutrophil-to-Lymphocyte Ratio
PLR	Platelet-to-Lymphocyte Ratio
MLR	Monocyte-to-Lymphocyte Ratio
CSF	Cerebrospinal Fluid
QAlb	CSF/Serum Albumin Quotient
QIgG	CSF/Blood Immunoglobulin G Quotient
QIgA	CSF/Blood Immunoglobulin A Quotient
QIgM	CSF/Blood Immunoglobulin M Quotient
ROC	Receiver Operating Characteristic
CDC	Complement-Dependent Cytotoxicity
CNS	Central Nervous System
IgG	Immunoglobulin G
ROC	Receiver Operating Characteristic
NMO-IgG	Immunoglobulin G Anti-Aquaporin-4 Autoantibodies

Declarations

Author Contributions

Xingyue Zheng: Conceptualization, Methodology, Formal analysis, Investigation, Data curation, Writing-original draft, Visualization.

Hongjing Yan: Conceptualization, Methodology, Writing-review & editing.

Hao Yin: Conceptualization, Data curation, Writing - review & editing.

Jing Shi: Conceptualization, Writing - review & editing.

Yuanyuan Liu: Conceptualization, Writing - review & editing.

Haotian Zhao: Conceptualization, Writing - review & editing.

Yuzhi Li: Conceptualization, Writing - review & editing.

Huakun Liu: Conceptualization, Writing - review & editing.

Lei Zhang: Conceptualization, Writing - review & editing.

ZhongruiYan: Conceptualization, Writing - review & editing, Supervision.

Chunbo Dong: Conceptualization, Methodology, Writing - review & editing, Supervision.

Consent for publication

Informed consent was obtained from each subject.

Competing interest

The authors declared no conflicts of interest.

References

1. Wingerchuk DM, Banwell B, Bennett JL, et al. International consensus diagnostic criteria for neuromyelitis optica spectrum disorders. Neurology. 2015. 85(2): 177-89.
2. Huang WC, Lin HC, Yang YH, et al. Neutrophil-to-lymphocyte ratio and monocyte-to-lymphocyte ratio are associated with a 2-year relapse in patients with multiple sclerosis. Mult Scler Relat Disord. 2022. 58: 103514.

3. Asavapanumas N, Verkman AS. Neuromyelitis optica pathology in rats following intraperitoneal injection of NMO-IgG and intracerebral needle injury. Acta Neuropathol Commun. 2014. 2: 48.

4. Bradl M, Lassmann H. Experimental models of neuromyelitis optica. Brain Pathol. 2014. 24(1): 74-82.

5. Orman G, Wang KY, Pekcevik Y, et al. Enhancing Brain Lesions during Acute Optic Neuritis and/or Longitudinally Extensive Transverse Myelitis May Portend a Higher Relapse Rate in Neuromyelitis Optica Spectrum Disorders. AJNR Am J Neuroradiol. 2017. 38(5): 949-953.

6. Shimizu F, Sano Y, Takahashi T, et al. Sera from neuromyelitis optica patients disrupt the blood-brain barrier. J Neurol Neurosurg Psychiatry. 2012. 83(3): 288-97.

7. Wang Y, Zhu M, Liu C, et al. Blood Brain Barrier Permeability Could Be a Biomarker to Predict Severity of Neuromyelitis Optica Spectrum Disorders: A Retrospective Analysis. Front Neurol. 2018. 9: 648.

8. Pierson ER, Wagner CA, Goverman JM. The contribution of neutrophils to CNS autoimmunity. Clin Immunol. 2018. 189: 23-28.

9. Yan SL, Russell J, Granger DN. Platelet activation and platelet-leukocyte aggregation elicited in experimental colitis are mediated by interleukin-6. Inflamm Bowel Dis. 2014. 20(2): 353-62.

10. Olsson A, Gustavsen S, Gisselø Lauridsen K, et al. Neutrophil-to-lymphocyte ratio and CRP as biomarkers in multiple sclerosis: A systematic review. Acta Neurol Scand. 2021. 143(6): 577-586.

11. Fu H, Qin B, Hu Z, et al. Neutrophil- and platelet-to-lymphocyte ratios are correlated with disease activity in rheumatoid arthritis. Clin Lab. 2015. 61(3-4): 269-73.

12. Song JE, Lee KY, Son GH. Prognostic Significance of Neutrophil-to-Lymphocyte Ratio for Repeat Cerclage in Women with Prolapsed Membranes. Biomed Res Int. 2018. 2018: 1507398.

13. Cabanillas-Lazo M, Cruzalegui-Bazán C, Pascual-Guevara M, et al. Clinical and imagenologic significance of the neutrophil-to-lymphocyte ratio in neuromyelitis optica spectrum disorder: A systematic review with meta-analysis. PLoS One. 2023. 18(2): e0281064.

14. Reiber H. Dynamics of brain-derived proteins in cerebrospinal fluid. Clin Chim Acta. 2001. 310(2): 173-86.

15. Kurtzke JF. Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). Neurology. 1983. 33(11): 1444-52.

16. Conradsson D, Ytterberg C, von Koch L, Johansson S. Changes in disability in people with multiple sclerosis: a 10-year prospective study. J Neurol. 2018. 265(1): 119-126.

17. Sicras-Mainar A, Ruíz-Beato E, Navarro-Artieda R, Maurino J. Impact on healthcare resource utilization of multiple sclerosis in Spain. BMC Health Serv Res. 2017. 17(1): 854.

18. Meixensberger S, Bechter K, Dersch R, et al. Sex difference in cerebrospinal fluid/blood albumin quotients in patients with schizophreniform and affective psychosis. Fluids Barriers CNS. 2020. 17(1): 67.

19. Chen B, Qin C, Tao R, et al. The clinical value of the albumin quotient in patients with neuromyelitis optica spectrum disorder. Mult Scler Relat Disord. 2020. 38: 101880.

20. Yuan C, Liu X, Cai S, et al. Secreted aminoacyl-tRNA synthetase-interacting multifunctional protein-1 (AIMP1) is a promising predictor for the severity of acute AQP4-IgG positive neuromyelitis optica spectrum disorder. Mult Scler Relat Disord. 2023. 70: 104504.

21. Ma L, Zeng A, Chen B, Chen Y, Zhou R. Neutrophil to lymphocyte ratio and platelet to lymphocyte ratio in patients with systemic lupus erythematosus and their correlation with activity: A meta-analysis. Int Immunopharmacol. 2019. 76: 105949.

22. Gasparyan AY, Ayvazyan L, Mukanova U, Yessirkepov M, Kitas GD. The Platelet-to-Lymphocyte Ratio as an Inflammatory Marker in Rheumatic Diseases. Ann Lab Med. 2019. 39(4): 345-357.

23. Carnero Contentti E, López PA, Criniti J, et al. Platelet-to-lymphocyte ratio differs between MS and NMOSD at disease onset and predict disability. Mult Scler Relat Disord. 2022. 58: 103507.

24. Chang BL, Ro LS, Chen CM, et al. Serum levels of cell adhesion molecules in patients with neuromyelitis optica spectrum disorder. Ann Clin Transl Neurol. 2020. 7(10): 1854-1861.

25. Yu Z, Fang X, Liu W, et al. Microglia Regulate Blood-Brain Barrier Integrity via MiR-126a-5p/MMP9 Axis during Inflammatory Demyelination. Adv Sci (Weinh). 2022. 9(24): e2105442.

26. Fu CC, Gao C, Zhang HH, et al. Serum molecular biomarkers in neuromyelitis optica and multiple sclerosis. Mult Scler Relat Disord. 2022. 59: 103527.
27. Sreeramkumar V, Adrover JM, Ballesteros I, et al. Neutrophils scan for activated platelets to initiate inflammation. Science. 2014. 346(6214): 1234-8.
28. Fang X, Sun S, Yang T, Liu X. Predictive role of blood-based indicators in neuromyelitis optica spectrum disorders. Front Neurosci. 2023. 17: 1097490.
29. Yan H, Wang Y, Li Y, et al. Combined platelet-to-lymphocyte ratio and blood-brain barrier biomarkers as indicators of disability in acute neuromyelitis optica spectrum disorder. Neurol Sci. 2024. 45(2): 709-718.
30. Sonia D'Souza C, Li Z, Luke Maxwell D, et al. Platelets Drive Inflammation and Target Gray Matter and the Retina in Autoimmune-Mediated Encephalomyelitis. J Neuropathol Exp Neurol. 2018. 77(7): 567-576.
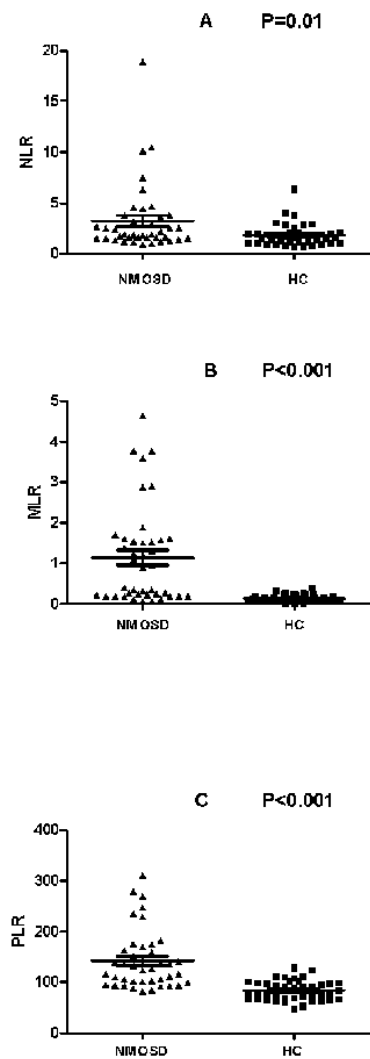
Figures

Figure 1

Comparison of serum inflammatory markers levels between NMOSD patients and HC patients.

A: Comparison of NLR levels between NMOSD patients and HC patients. NLR: Neutrophil-to-lymphocyte ratio.

B: Comparison of MLR levels between NMOSD patients and HC patients. MLR: Monocyte-to-lymphocyte ratio.

C: Comparison of PLR levels between NMOSD patients and HC patients. PLR: Platelet-to-lymphocyte ratio.
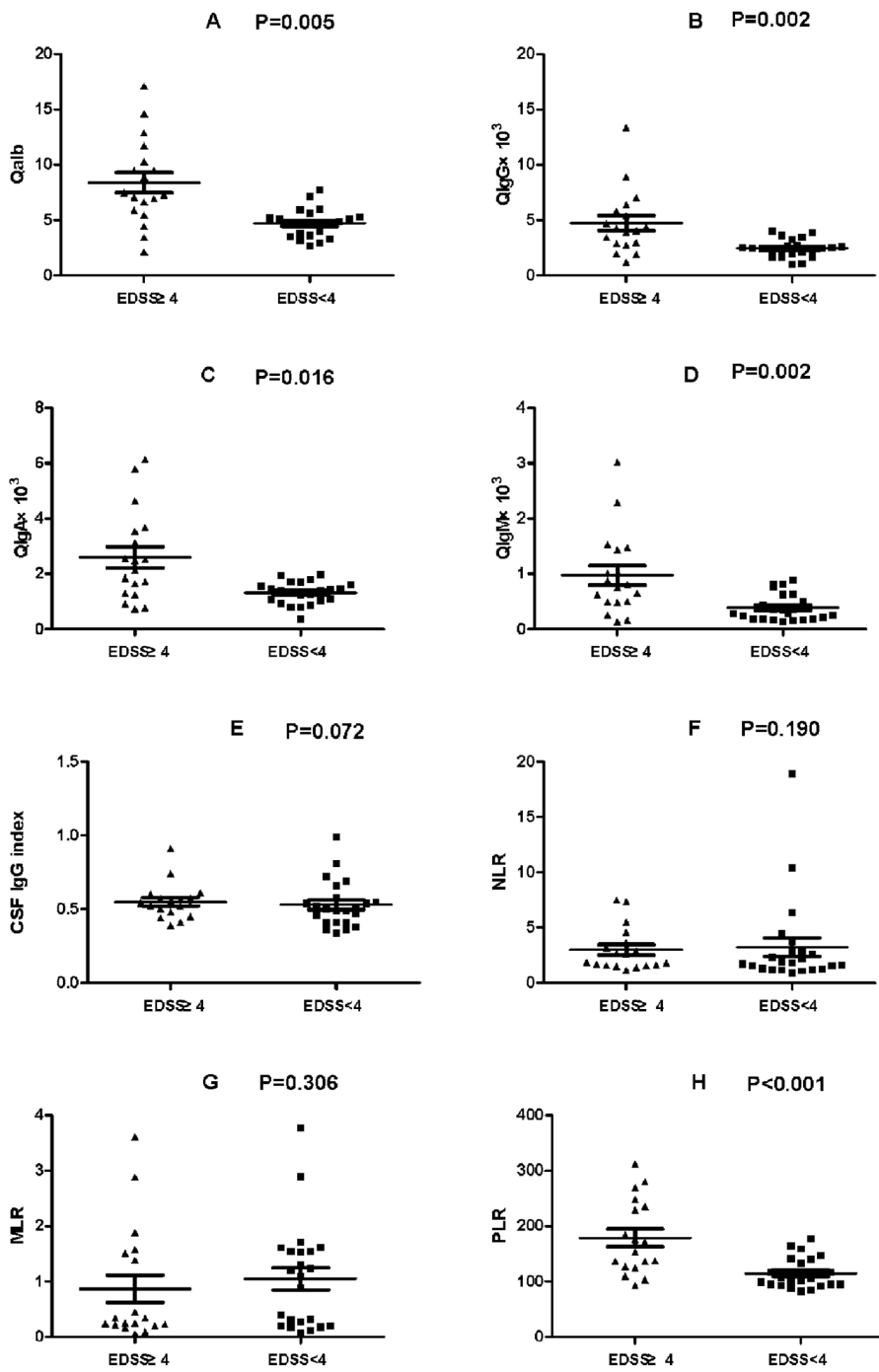
Figure 2

Comparison of various indicators between the EDSS<4 group and the EDSS≥4 group.

A: QAlb levels are higher in the EDSS≥4 groups compared to the EDSS<4 group.

B: QIgG levels are higher in the EDSS≥4 groups compared to the EDSS<4 group.

C: QIgA levels are higher in the EDSS≥4 groups compared to the EDSS<4 group.

D: QIgM levels are higher in the EDSS≥4 groups compared to the EDSS<4 group.

E: Comparison of cerebrospinal fluid IgG index between the EDSS<4 group and the EDSS≥4 group.

F: Comparison of NLR between the EDSS<4 group and the EDSS≥4 group.

G: Comparison of MLR between the EDSS<4 group and the EDSS≥4 group.

H: Comparison of PLR between the EDSS<4 group and the EDSS≥4 group.

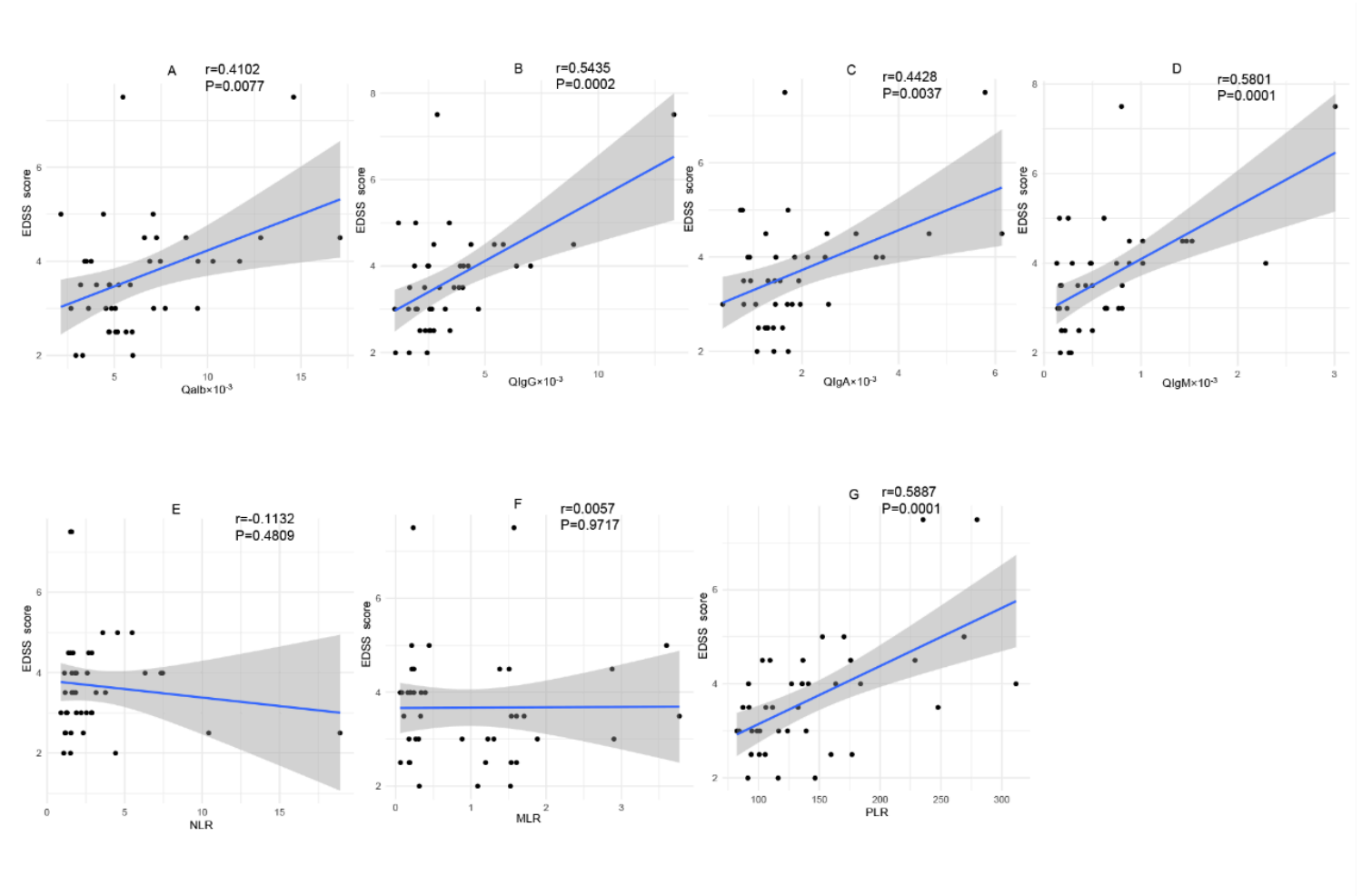
Figure 3

A-G Spearman rank correlation between EDSS score and indicators.

A: The increase of QAlb was positively correlated with EDSS score (r=0.4102， P=0.0077).

B: The increase of QIgG was positively correlated with the EDSS score(r=0.5435， P=0.0002);

C: The increase of QIgA was positively correlated with the EDSS score (r=0.4428， P=0.0037);

D: The increase of QIgM was positively correlated with the EDSS score (r=0.5801， P=0.0001);

E: Correlation between EDSS score and NLR (r=-0.1132，P=0.4809);

F: Correlation between EDSS score and MLR(r=0.0057，P=0.9717);

G: The increase of PLR was positively correlated with EDSS score r=0.5887， P=0.0001);

EDSS, Expanded Disability Status Scale; NLR, neutrophil-to-lymphocyte ratio; MLR, lymphocyte to monocyte ratio; PLR, platelet count to lymphocyte ratio; QAlb, albumin quotient; QIgG, CSF/serum total IgG ratio; QIgM, CSF/serum total IgM ratio; QIgA, CSF/serum total IgA ratio.
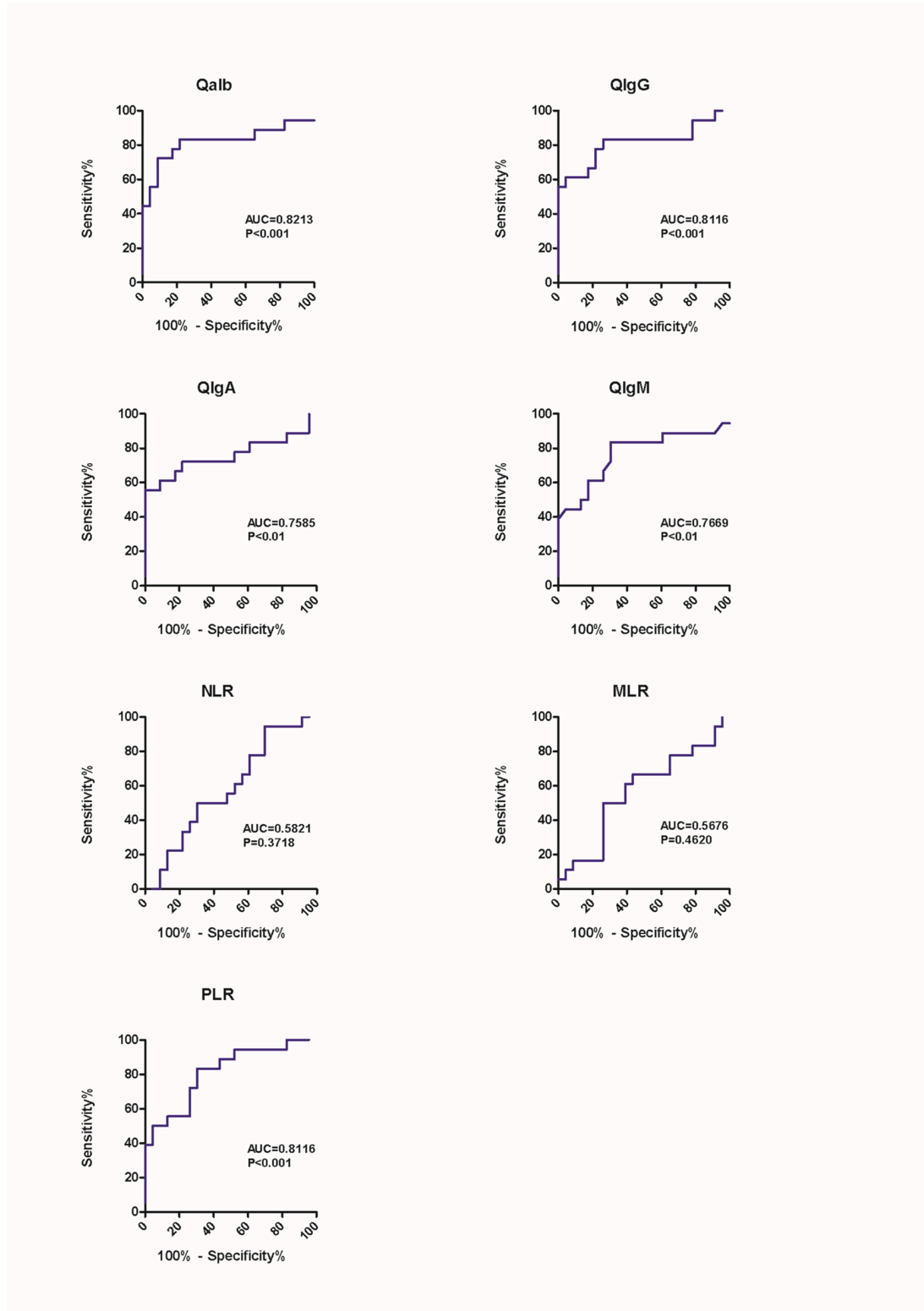

Figure 4

Blood-brain barrier and PLR biomarkers can predict the severity of acute NMOSD.

ROC curve analysis was used to distinguish between acute NMOSD patients with EDSS score≥4 and those with EDSS score<4 using QAlb, QIgG, QIgA, QIgM, NLR, MLR, and PLR. Optimal cut-off values for QAlb, QIgG, QIgA, QIgM, and PLR levels were determined

based on the ROC curve area to predict acute NMOSD with EDSS≥4 accurately. The optimal cut-off values for QAlb, QIgG, QIgA, and PLR show good specificity and sensitivity in predicting disability severity in patients based on EDSS scores, making them clinically valuable for prediction. NLR and MLR do not have a significant correlation with EDSS scores, showing no clinical predictive value.

EDSS, Expanded Disability Status Scale; NLR, neutrophil-to-lymphocyte ratio; MLR, lymphocyte to monocyte ratio; PLR, platelet count to lymphocyte ratio; QAlb, albumin quotient; QIgG, CSF/serum total IgG ratio; QIgM, CSF/serum total IgM ratio; QIgA, CSF/serum total IgA ratio.