INDONESIAN JOURNAL OF
CLINICAL PATHOLOGY AND MEDICAL LABORATORY
Majalah Patologi Klinik Indonesia dan Laboratorium Medik

PAGE 291
2022 July; 28(3): 291-296
p-ISSN 0854-4263 e-ISSN 2477-4685
Available at www.indonesianjournalofclinicalpathology.org

Risks of Hemorrhage and Poor Clinical Outcome in SLE with Thrombocytopenia at Dr. Sardjito Hospital

Surawijaya Bakhtiar Kaslam, Umi Solekhah Intansari

Department of Clinical Pathology and Laboratory Medicine, Faculty of Medicine, Nursing and Public Health, Gadjah Mada University, Yogyakarta, Indonesia. E-mail: zoera_ku2000@yahoo.com

ABSTRACT

Systemic Lupus Erythematosus (SLE) is an autoimmune disease that affects various body organs and causes chronic inflammation. Thrombocytopenia is common in SLE, and there is a risk of causing bleeding, which can result in death. This study aimed to analyze the relationship of thrombocytopenia with bleeding and poor clinical outcomes in SLE patients at Dr. Sardjito Hospital. The design of this study was retrospective observational analytic. The research subjects were patients diagnosed with SLE at Dr. Sardjito Hospital from January 2016-December 2019 who conducted ANA and anti-dsDNA examinations. Statistical analysis using MedCalc version 13.0. Receiver operating characteristic curve analysis to determine the cut-off value of the platelet count for the occurrence of bleeding. Chi-Square for trend test to determine the relationship between the degree of thrombocytopenia and the degree of bleeding. Kaplan-Meier test to determine the six months survival analysis for SLE patients. There were 61 SLE patients at Dr. Sardjito Hospital. Thirty-two patients (52.5%) had thrombocytopenia. The AUC of the platelet count for the occurrence of hemorrhage was 0.988 (95% CI=0.918-1, $p < 0.0001$), the cut-off value was 146x103/L, sensitivity 100%, specificity 90.6%, and LR+ 10.33. The AUC of the platelet count for grade 3 hemorrhage was 0.929 (95% CI=0.833-0.979, $p < 0.0001$), cut-off value 91x103/L, sensitivity 100%, specificity 89.3%, and LR+ 9.33. Hemorrhage was seen in 29 subjects with thrombocytopenia. Five subjects (8.2%) died, with a significant difference in the mortality of SLE patients with and without thrombocytopenia in the six months survival analysis (p=0.028). The risk of hemorrhage and poor clinical outcome (death) were significantly higher in SLE patients with thrombocytopenia and increased with the thrombocytopenia grade.

Keywords: Thrombocytopenia, hemorrhage, systemic lupus erythematosus

INTRODUCTION

Systemic Lupus Erythematosus (SLE) is an autoimmune disease that affects multiple organs and can cause chronic inflammation. Systemic lupus erythematosus shows a wide spectrum of clinical manifestations and is related to various autoantibodies.[1] Factors such as genetics, epigenetics, hormonal, environmental, and immunoregulatory are involved in the expression of this disease.[2] Clinical manifestations of this disease are various such as polyarthritis, oral ulcer, rashes, and hematological, kidney and brain disorders. Systemic lupus erythematosus usually happens in females with a ratio of 12:1, and death is usually caused by kidney failure.[3] Hemorrhage complications that involve multiple systems can also occur in SLE, but the exact mechanism is still not fully understood.[4]

Hematological abnormalities, such as thrombocytopenia and leukopenia, are clinical manifestations often found in SLE. Thrombocytopenia is known as one of the hematological criteria according to the American College of Rheumatology (ACR) classification. Thrombocytopenia often happens in SLE and is a risk factor for bleeding. The prevalence is around 10–40%, but severe thrombocytopenia rarely occurs.[1]

There is a significant increase in hemorrhage risks found in SLE patients with mild thrombocytopenia. In contrast, SLE patients with severe thrombocytopenia often experience bleeding, showing that thrombocytopenia is a vital prognosis factor that is harmful.[5] Abdel Galil *et al.* state that thrombocytopenia is related to SLE activity and is strongly associated with arthritis, nerve involvement, and lupus nephritis. The platelet count shows a reverse correlation with SLE activity.[6]

Thrombocytopenia is related to severe clinical manifestations of SLE, such as neuropsychiatric symptoms, kidney involvement, and hemolytic anemia. Furthermore, thrombocytopenia is also known to be related to the prognosis of SLE, including death.[1] This study analyzed the

relationship between thrombocytopenia with hemorrhage and poor clinical outcomes in SLE patients at Dr. Sardjito Hospital, Yogyakarta.

METHODS

This study was an analytic observational retrospective by seeing the data from the medical records of patients diagnosed with SLE in Dr. Sardjito Hospital and collecting the patients' primary data, clinical data, laboratory examination data, and other supporting tests. The diagnosis of SLE is made according to the criteria of the American College of Rheumatology (ACR) and ANA and anti-dsDNA examinations. The subjects of this study are patients diagnosed with SLE in Dr. Sardjito Hospital from January 2016–December 2019. Exclusion criteria were patients with SLE that didn't have complete clinical and laboratory data.

Patients'laboratory examinations were complete blood count, Blood Urea Nitrogen (BUN), creatinine, urinalysis, ANA, and anti-dsDNA. An EDTA blood sample for complete blood count is taken on the same day as the serum sample. The method used to examine the complete blood count was with an automatic instrument, namely the Sysmex XN1000–Sysmex XN550 (flow cytometry method) or Advia 120 (optical method). Even though the procedures of these three instruments are different, they correlate well with routine hematological examinations. The attending residents confirmed the results of platelet counts that were < $100x10^3$/dL manually. Blood urea nitrogen and creatinine were measured using the Cobas e601 and Dimension ex12000, using the same examination principles. Urinalysis was done using the UF500i.

Thrombocytopenia was classified as mild thrombocytopenia (platelet count 100–$149x10^3$/L), moderate thrombocytopenia (platelet count 50–$99x10^3$/L) and severe thrombocytopenia (platelet count <50x103/L).[7] Method of hemorrhage classification adopted the criteria by Ziakas *et al.* and Massachusetts Medical Center, where the degrees were divided into grade 0 (no hemorrhage found), grade 1 (mucocutaneous hemorrhage, ecchymosis near injection site, hemorrhage spots in the skin or purpura), grade 2 (obvious hemorrhage such as hemorrhage from the nose, in the urine, from the genitals or gut system) and grade 3 (hemorrhage that needs more than one unit of red blood cells a day for transfusion, such as life-threatening hemorrhage, intracranial and respiratory hemorrhage).[5] Disease activity on the patient's admission was measured by SLE Disease Activity Index (SLEDAI). The SLEDAI score interpretation is as follows: no activity (SLEDAI=0), mild activity (SLEDAI=1–5), moderate activity (SLEDAI=6–10), high activity (SLEDAI=11–19), and very high activity (SLEDAI > 20).[8]

Statistical analysis used MedCalc version 13.0 with $p<0.05$ deemed as statistically significant. Data distribution was analyzed using Shapiro-Wilk. Parametric data were presented as mean+Standard Deviation (SD). Non-parametric data were presented as median (minimal–maximal). Categorical data were presented in proportion tables. The receiver operating characteristic analysis curve was done to determine the platelet count's cut-off value (Youden index) that would cause hemorrhage in SLE patients. Chi-Square for trend test was used to find the relationship between thrombocytopenia grade with the degree of bleeding in research subjects. Kaplan-Meier was implemented to find the six months survival rate of SLE patients.

The study was approved by Ethic Committee Gadjah Mada University/Dr. Sardjito Hospital with number KE/FK/0079/EC/2021.

RESULTS AND DISCUSSIONS

Sixty-one SLE patients at Dr. Sardjito Hospital during the 2016–2019 period. As stated in Table 1, study subjects were dominated by females (95.1%), with ages mostly > 18 years old (60.7%). There were 32

Table 1. Characteristics of study subjects

Parameter	Total (%)
Age	
< 18 years old	24 (39.3%)
≥ 18 years old	37 (60.7%)
Gender	
Male	3 (4.9%)
Female	58 (95.1%)
Platelet count	
< 50 x 10^3/μL	5 (8.2%)
50-99 x 10^3/μL	8 (13.1%)
100-149 x 10^3/μL	19 (31.2%)
≥ 150 x 10^3/μL	29 (47.5%)
Hemorrhage	
Grade 0	32 (52.4%)
Grade 1	7 (11.5%)
Grade 2	17 (27.9%)
Grade 3	5 (8.2%)
SLEDAI score	
0 (no activity)	0 (0%)
1–5 (mild activity)	0 (0%)
6–10 (moderate activity)	0 (0%)
11–19 (high activity)	34 (55.7%)
≥ 20 (very high activity)	27 (44.3%)
Patient outcome	
Alive	56 (91.8%)
Dead	5 (8.2%)

Table 2. Clinical characteristics dan laboratory results of the research subjects

Parameter	Total (n=61)	With Thrombocytopenia (n=32)	Without Thrombocytopenia (n=29)	p
Hemoglobin (g/dL)	10.35±1.93	9.32±1.68	11.49±1.52	**<0.0001**[a]
Erythrocytes (x10^6/µL)	3.86±0.81	3.45±0.81	4.3±0.55	**<0.0001**[a]
Hematocrit (%)	31.39±5.89	28.15±5.17	34.97±4.41	**<0.0001**[a]
Leukocytes (x10^3/µL)	5.87 (1.46–29.88)	4.81 (1.46-29.88)	6.27 (2.71-22.22)	**0.028**[b]
BUN (mg/dL)	11.4 (5-97.2)	14.95 (5-97.2)	10.7 (6.8-37)	**0.033**[b]
Creatinine (mg/dL)	0.66 (0.29-5.76)	0.69 (0.29-5.76)	0.65 (0.4-2.82)	0.402[b]
Anti-dsDNA (U/mL)	200 (25.4-200)	200 (25.4-200)	70.9 (25.4-200)	**0.01**[b]
ANA				0.796[c]
Positive, n (%)	42 (68.9%)	23 (71.9%)	19 (65.5%)	
Negative, n (%)	19 (31.1%)	9 (28.1%)	10 (34.5%)	
Hemorrhage				**<0.0001**[d]
Grade 0, n (%)	32 (52.4%)	3 (9.4%)	29 (100%)	
Grade 1, n (%)	7 (11.5%)	7 (21.9%)	0 (0%)	
Grade 2, n (%)	17 (27.9%)	17 (53.1%)	0 (0%)	
Grade 3, n (%)	5 (8.2%)	5 (15.6%)	0 (0%)	
SLEDAI score				**0.025**[c]
0, n (%)	0 (0%)	0 (0%)	0 (0%)	
1-5, n (%)	0 (0%)	0 (0%)	0 (0%)	
6-10, n (%)	0 (0%)	0 (0%)	0 (0%)	
11-19, n (%)	34 (55.7%)	13 (40.6%)	21 (72.4%)	
≥20, n (%)	27 (44.3%)	19 (59.4%)	8 (27.6%)	
Patients outcome				0.079[c]
Alive, n (%)	56 (91.8%)	27 (84.4%)	29 (100%)	
Death, n (%)	5 (8.2%)	5 (15.6%)	0 (0%)	

[a]Analysis results of independent T- test between patients with and without thrombocytopenia
[b]Analysis results of Mann-Whitney test between patients with and without thrombocytopenia
[c]Analysis results of Chi-Square test between patients with and without thrombocytopenia
[d]Analysis results of Chi-Square for trend test between patients with and without thrombocytopenia

patients (52.5%) that experienced thrombocytopenia with the platelet count < 150x10^3/L, with three males and 29 females (age median at 18.5 years old, with a range of 10–56 years old). The youngest patient was ten years old. This result is in line with an 8-year study by Laustrup *et al.* stated that young active SLE patients were more likely to have thrombocytopenia.[5] This research also had 29 patients that experienced hemorrhage with various grades, and five patients that had been observed for six months stated having SLE passed away. The patient's disease activity was calculated on admission by the SLEDAI score, and the results show that 34 patients were in high activity and 27 patients were in a very high activity state.

Table 2 shows these research subjects' clinical characteristics and laboratory results, divided into two groups: thrombocytopenia and without thrombocytopenia. Data distribution of the subjects were analyzed using Shapiro-Wilk and each parameter was analyzed for its relation with the thrombocytopenia and without thrombocytopenia group. The data accumulated showed a statistically significant difference ($p<0.05$) in the following parameters: Hemoglobin, erythrocyte, leukocyte, BUN, anti-dsDNA, hemorrhage, and SLEDAI score.

The receiver operating characteristic curve analysis showed in Figure 1 (A) to determine the platelet count cut-off (Youden index) that caused a hemorrhage in SLE patients. Area under ROC curve for the platelet count was 0.988 (95% CI=0.918-1, $p <0.0001$). The cut-off value for the platelet count that caused hemorrhage was 146x10^3/L, with a sensitivity of 100%, specificity of 90.6%, and a positive likelihood ratio of 10.33.

Figure 1 (B) shows the ROC curve of platelet count that causes a grade 3 hemorrhage in SLE patients, with an area under the ROC curve for platelet count was 0.929 (95% CI=0.833–0.979, $p<0.0001$). The cut-off for platelet count that causes a grade 3 hemorrhage was 91x10^3/L with a sensitivity of 100% and specificity of 89.3%, and a positive likelihood ratio 9.33.

Table 3 explains the relationship of the degree of thrombocytopenia with the grade of hemorrhage in the research subjects. Hemorrhage was found in 29 of

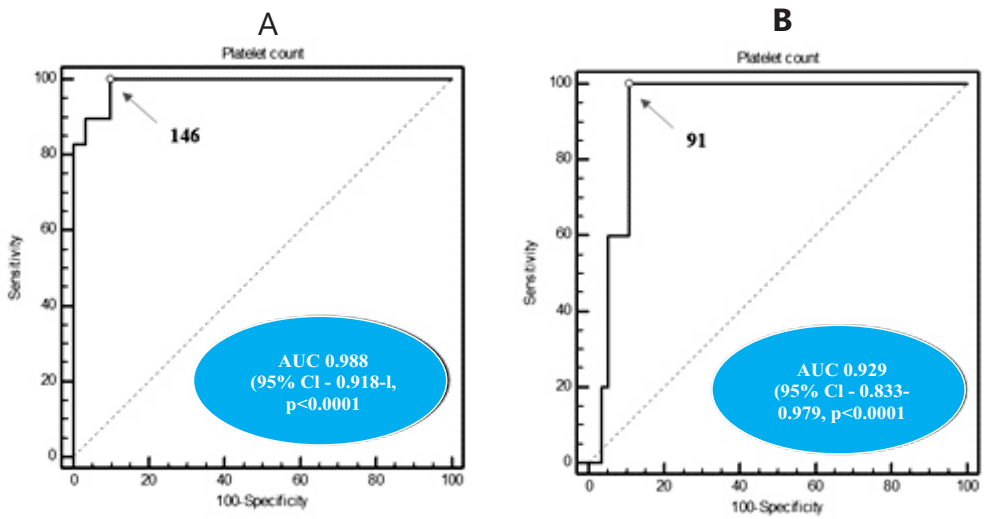

Figure 1 (A). The receiver operating characteristic curve of platelet count experienced bleeding (B). Receiver operating characteristic curve for platelet count that experienced Grade 3 hemorrhage

Table 3. The relationship of the degree of thrombocytopenia with the grade of hemorrhage

Thrombocytopenia	Hemorrhage				TOTAL	p
	Grade 0	Grade 1	Grade 2	Grade 3		
Mild	3	5	11	0	19 (59.4%)	**0.019***
Moderate-severe	0	2	6	5	13 (40.6%)	
TOTAL	3	7	17	5	32	
	(9.4%)	(21.9%)	(53.1%)	(15.6%)		

* Analysis result of Chi-Square for trend test between the degree of thrombocytopenia and grade of hemorrhage

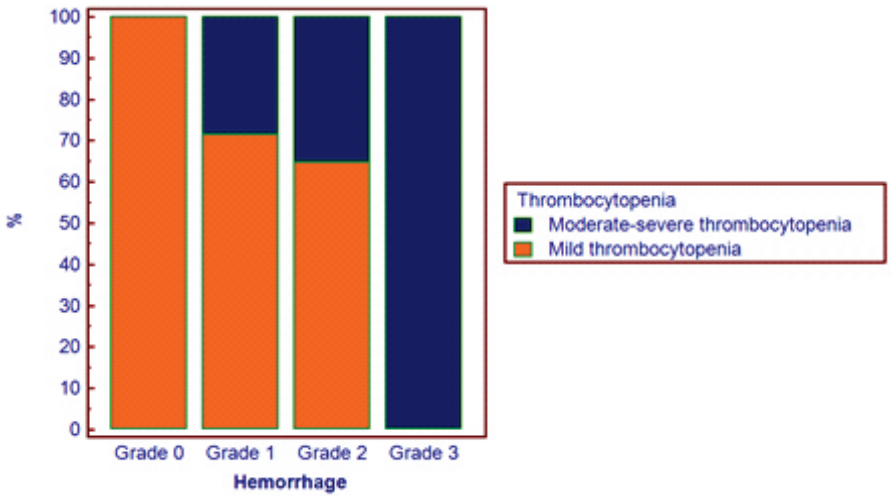

Figure 2. Percentage of the degree of thrombocytopenia in each degree of hemorrhage

32 subjects with thrombocytopenia, divided into 19 subjects (59.4%) with mild thrombocytopenia, 8 subjects (25%) with moderate thrombocytopenia and five subjects (15.6%) with severe thrombocytopenia. There were only a few subjects with moderate and severe thrombocytopenia, so they were merged into moderate-severe thrombocytopenia.

There were grade 2 hemorrhages that happened the most (gum hemorrhage, hematuria, melena), 53.1%. There were only 3 subjects that had thrombocytopenia without hemorrhage (9.4%). Thrombocytopenia in SLE is linked with lupus nephritis, neuropsychiatry, pericarditis, leukopenia, infection, activation of lupus, etc.[5] In this research, most cases with complication of lupus nephritis happened in the mild thrombocytopenia group that manifested into hematuria. This finding explains why grade 2 hemorrhage occurs even though the patient only had

mild thrombocytopenia. Hemorrhage risks increase as thrombocytopenia worsens accordingly to the grade of SLE. This fact is in line with Li *et al.*, which compared the hemorrhage risks in SLE patients with moderate and severe thrombocytopenia.[5] This study also found that grade 3 hemorrhage that is life-threatening happened more in moderate-severe thrombocytopenia, with statistically significant results (p=0.019).

The percentage of the degree of thrombocytopenia in each degree of hemorrhage is presented in Figure 2. In grade 0 (without hemorrhage) there was only mild thrombocytopenia (platelet count 100–149x10^3/L). In grade 1 hemorrhage (ecchymosis, petechiae, purpura) and grade 2 (hematemesis, melena, hematuria, epistaxis, gum bleeding) was dominated by mild thrombocytopenia even though it was followed by moderate-severe thrombocytopenia proportion. Grade 3 hemorrhage (intracranial hemorrhage, hemoptoe) was only found in conditions of moderate-severe thrombocytopenia (platelet count < 100x10^3/L).

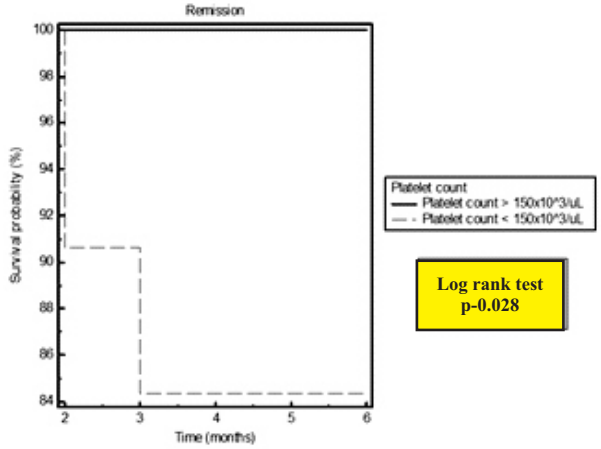

Figure 3. Six months survival analysis of SLE patients

This research showed five subjects passed away (8.2%). As seen in the six months survival analysis in Figure 3, the mortality of SLE patients with thrombocytopenia was significantly different compared to patients that didn't have thrombocytopenia with log-rank test p=0.028.

Thrombocytopenia is a common clinical manifestation in SLE and has been reported to have a strong relationship with serious clinical manifestations. The low levels of complements and increment of anti-dsDNA antibody have also been observed in patients with thrombocytopenia.[1] Thrombocytopenia indicates a poor SLE prognosis. Thrombocytopenia is an independent risk factor for death and has been found to be linked with higher disease activity and more significant damage in different groups of SLE. Thrombocytopenia is linked with more severe disease and negatively affects an SLE patient's well-being. Thrombocytopenia has been proven to be a predictor in patients with higher disease activity, the possibility of organ destruction, and death.[9] In this study, patients with a platelet count of 146x10^3/L, have a 10.33 risk of bleeding, while patients with a platelet count of 91x10^3/L have a 9.33 chance of grade 3 hemorrhage that is life-threatening. The death level of patients with thrombocytopenia is 24%, significantly higher than SLE patients without thrombocytopenia. The degree of thrombocytopenia also affects the welfare of an SLE patients' life, whether they have other severe clinical manifestations, abnormal laboratory results, and SLE disease activity.[1]

Severe thrombocytopenia in SLE seldom happens but a potentially massive hemorrhage (intracranial, intraperitoneal) could be life-threatening. Thrombocytopenia, which happens to most patients with SLE, is caused by an increase in immune-mediated thrombocyte destruction in the peripheral circulation. This study shows that antiphospholipid antibody (aPL) has a strong correlation with thrombocytopenia. This antibody work against cardiolipin, the phospholipid of the thrombocytes wall, and phospholipid component from the prothrombin activator complex. Anticardiolipin antibody (ACL) is one of the many antiphospholipid antibodies that are considered to have a higher risk of thrombocytopenia happening in an SLE patient compared to aPL antibody that is well known to be a lupus anticoagulant.[10]

Limitations in this study are that the disease activity of SLE when the patient was admitted that was calculated using the SLEDAI score only showed high activity and very high activity, so a whole description of the relationship of various SLE activities with the clinical manifestations that happen could not be calculated. In addition, the history of medication that the patient got in prior health facilities was not documented, so the disease course could not be observed comprehensively.

CONCLUSIONS AND SUGGESTIONS

Hemorrhage risks and poor outcome (death) are significantly higher in SLE patients with thrombocytopenia and increase with the degree of thrombocytopenia. Further research is needed with a larger sample to get important data on the relationship of thrombocytopenia in SLE with

hemorrhage and the poor outcomes that happens. One must be cautious of the risk of hemorrhage and poor outcomes in patients with severe SLE and thrombocytopenia.

REFERENCES

1. Jung JH, Soh MS, Ahn YH, Um YJ, Jung JY, *et al*. Thrombocytopenia in systemic lupus erythematosus: Clinical manifestations, treatment, and prognosis in 230 patients. Medicine, 2016; 95(6): e2818.
2. Pamuk ON, Balci MA, Donmez S, Tsokos GC. The incidence and prevalence of systemic lupus erythematosus in Thrace, 2003-2014: A-12-year epidemiological study. Lupus, 2016; 25: 102–109.
3. Wijaya C, Nurulita A, Bahrun U. Correlation of antinuclear antibody (ANA) profile with hematologic and renal disorders in systemic lupus erythematosus. Indonesian Journal of Clinical Pathology and Medical Laboratory, 2017; 23(2): 146–150.
4. Abdulla MC. Spontaneous soft tissue hemorrhage in systemic lupus erythematosus. Rheumatism, 2016; 68(4): 199-202.
5. Li J, Pan Z, Liu H, Ding F, Shu Q, Li X. Retrospective analysis of the risk of hemorrhage associated with moderate and severe thrombocytopenia of 173 patients with systemic lupus erythematosus. Medicine, 2018; 97: 27.
6. Abdel Galil SM, Edrees AM, Ajeeb AK, Aldoobi GS, Boshy ME, Hussain W. Prognostic significance of platelet count in SLE patients. Platelets, 2017; 28: 203-207.
7. Nagrebetsky A, Samkari HA, Davis NM, Kuter DJ, Kronish JPW. Perioperative thrombocytopenia: Evidence, evaluation, and emerging therapies. British Journal of Anaesthesia, 2019; 122(1): 19-31.
8. Sadiq AM, Assey EV, Sadiq AM, Dekker MCJ, Howlett WP. Fatal cerebral venous sinus thrombosis as a manifestation of uncontrolled systemic lupus erythematosus in a young African female. Clin Case Rep, 2021; 9: e04454.
9. Zhu FX, Huang JY, Ye Z, Wen QQ, Wei JCC. Risk of systemic lupus erythematosus in patients with idiopathic thrombocytopenic purpura: A population-based cohort study. Ann Rheum Dis, 2020; 0: 1–7.
10. Zhang J, Chen Y, Yu Z, Liu L. Bilateral hemorrhagic retinopathy with Roth spots in pediatric-onset systemic lupus erythematosus and associated thrombocytopenia: A case report and review of literature. Ocular Immunology & Inflammation, 2017; 00(00): 1–4.