OLGU SUNUMU / *CASE REPORT*

A Review on Differential Diagnosis and Diagnostic Criteria of Complement-Mediated Thrombotic Microangiopathy with a PLASMIC Score Below Six and Coexisting Hepatitis B Positivity in a Male Patient

PLASMIC Skoru Altı Olan ve Hepatit B Pozitifliğinin Eşlik Ettiği Kompleman Aracılı Trombotik Mikroanjiyopati Saptanan Bir Erkek Hasta: Ayırıcı Tanı ve Tanı Kriterleri Üzerine Bir İnceleme

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ABSTRACT

Microangiopathic Hemolytic Anemia (MAHA); Congenital Thrombotic thrombocytopenic purpura (TTP), Acquired (Immune) TTP, Shiga toxin associated Endemic hemolytic uremic syndrome (HUS) and Complement-Mediated TMA (CM-TMA), which may present with different clinical findings, Thrombocytopenia is a severe condition that affects multiple organ systems with anemia. In the congenital form, ADAMTS13 (von Willebrand Factor-Cleaving Protease or a metalloprotease that belongs to the "α disintegrin and metalloprotease with a thrombospondin type I motif) is diagnosed by the deficiency of the enzyme and the absence of antibodies. While in autoimmune TTP, the enzyme deficiency is associated with antibodies, endemic HUS associated with Shiga toxin is characterized by decreased ADAMTS13 levels due to endothelial damage. CM-TMA is associated with complement factor H (CFH) inhibitory dysfunction and increased complement levels due to a genetic mutation. On March 27, 2023, the patient with complaints of shortness of breath, headache, dizziness, weakness, and numbness in the hands and arms was admitted to the internal medicine clinic.

The patient, presenting with Thrombocytopenia, reduced haptoglobin levels, elevated reticulocyte count, increased LDH, indirect hyperbilirubinemia, and a PLASMIC score of 6 in peripheral blood smear, was hospitalized and treated with the prediagnosis of TTP. Later, ADAMTS13 level was found to be 73% (normal range: 40–130), and the diagnosis of CM-TMA was considered. In addition, we discussed the clinical distinction and treatment of TTP by reviewing the literature.

Keywords: *microangiopathic hemolytic anemia, thrombotic thrombocytopenic purpura, complement-related thrombotic thrombocytopenic purpura, ADAMTS13 protein, schistocyte, plasmic score*

ÖZET

Mikroanjiyopatik Hemolitik Anemi (MAHA); Konjenital Trombotik Trombositopenik Purpura (TTP), Edinsel (Otoimmün) TTP, shiga toksin ile ilişkili Endemik hemolitik üremik sendrom (HÜS) ve Complement-Mediated TMA (CM-TMA) olarak farklı klinik bulgular ile ortaya çıkabilen, trombositopeni, anemi ile seyreden birden çok organ sistemini etkileyen ciddi bir tablodur. Konjentinal formunda ADAMTS13 (Von Willebrand Faktör Ayırıcı Proteaz veya disintegrin'e ait bir metaloproteaz ve trombospondin tip I motifli metalloproteaz) olarak adlandırılan enzimin eksikliği ve antikor yokluğu ile teşhis edilirken, otoimmün TTP de ise enzim eksikliği antikorların varlığıyla ilişkilidir. Shiga toksini ile ilişkili endemik HÜS ise oluşan endotel hasarı sonucunda ADAMTS13 düzeylerinin azalması ile karekterezidir. CM-TMA ise bir genetik mutasyon sonucunda Complement Factor H (CFH) inhibitör fonksiyon bozukluğu ve kompleman düzeylerinin artması ile ilişkilidir. 27 mart 2023 tarihinde nefes darlığı, baş ağrısı, baş dönmesi, halsizlik, el ve kollarda uyuşma şikâyetleri olan 40 yaşındaki erkek hasta iç hastalıkları kliniğine yatırıldı.

Trombositopeni, haptoglobulin düşüklüğü, retikülositte artma, LDH yüksekliği, endirekt hiperbillüribinemi ve çevre kan yaymasında şistositleri görülen ve PLASMIC skoru 6 olan hasta TTP ön tanısı ile hastaneye yatırılarak tedavisi düzenlendi. Daha sonra ADAMTS13 düzeyi %73 (normal aralık: 40–130) saptanan hastada CM-TMA tanısı düşünüldü. Bununla beraber literatür taranarak TTP'nın klinik ayrımı ve tedavisine değindik

Anahtar kelimeler: *mikroanjiyopatik hemolitik anemi, trombotik trombositopenik purpura, kompleman-ilişkili trombotik trombositopenik purpura, ADAMTS13 protein, şistosit, plazmik skor*

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Introduction

Trombotik trombositopenik purpura is a disease that affects 1–5 people out of 1 million people in the population and has a high mortality rate. Antibodies against ADAMSTS13 constitute 2/3 of cases, either congenital or acquired and/or autoimmune. Trombotik trombositopenik purpura is a microangiopathy syndrome caused by congenital or acquired deficiency of ADAMTS13, which breaks down the von Willebrand factor (VWF). in the Table 1, either the function of ADAMSTS13 metalloprotease is impaired or its level decreases. Shiga toxin-mediated hemolytic uremic syndrome (ST-HUS), CM-TMA (Atypical HUS), drug-induced and other rare inherited TMA syndromes[1].

Therefore, to improve this situation, plasmapheresis (PLEX), fresh frozen plasma (FFP), and Rituximab (CD20 inhibitor) are started. Rituximab has a significant clinical effect as a salvage therapy for relapse. As a biosynthetic human-mouse chimeric monoclonal antibody, rituximab can reduce B lymphocytes with an abnormal immune response to the antigen[2]. In particular, volume load and blood-borne infectious agents should not be ignored in applying FFP. In the case of CM-TMA, this deadly disease can be prevented by administering eculizumab (C5 inhibitor)[3]. TTP, HUS, MAHA, and Thrombocytopenia are partially similar acute syndromes affecting multiple organ systems. Among the main diagnostic features, the most important is microangiopathic hemolytic anemia, characterized by non-immune hemolysis, negative direct Coombs test, and erythrocyte fragmentation in peripheral blood. As typical hemolysis findings, an Increase in serum indirect bilirubin and LDH levels (due to tissue damage and hemolysis) is observed. In the peripheral blood smear of patients with TTP-HUS syndrome, erythrocyte fragmentation is found in an average of 8% (between 1–18%) of erythrocytes. Trombotik trombositopenik purpura Pentad, MAHA, Thrombocytopenia, renal failure, fever, and neurological symptoms are found. The platelet count is around 25×109/l (5–120×109/). Neurological symptoms may be in the form of confusion or severe headache, as well as transient ischemic attack, seizure, and coma. Although fever cases are reported less frequently, when detected, it may indicate a septic Picture[4,5].

Case Presentation

A 40-year-old male patient, 85 kilos and 1.85 meters tall, applied to our internal medicine clinic with complaints of headache, dizziness, confusion, numbness in the hands and arms, and weakness. In his first examination, blood pressure was 110/70 mm/Hg; pulse was 88/min; fever was 38°C. The respiratory rate was 18/min, the head and neck examination was unremarkable, the respiratory and cardiovascular system examination was unremarkable, and no purpura or petechiae were found on his skin. The blood biochemistry was as follows: Lactate Dehydrogenase (LDH) 1345 U/L (0–248), ferritin 875 (24–336 ng/mL, for

Table 1. *The patient's biochemical, complete blood count, and some hormone analysis results*

Laboratory parameters and normally value	First day	Fifth day	After one week	After one month
ADAMTS13 level (40–130%)	73%	(4 months after treatment)		
Leukocyte (3700–10400 cells/microL)	8100	7012	7482	8500
Neutrophil (40–75%)	51	74	76	70
Lymphocytes (16–49%)	35	23	14.9	23.8
Hemoglobin (HGB)(g/dL)	**6.4**	**9.1**	**9.9**	**14.2**
MCV (78–98 fL)	**100**	**107**	**110**	**98**
Creatinine (0.67–1.17 mg/dL)	**1.35**	**0.80**	**0.82**	**0.83**
PLT (149–371×10^3 cells/microL)	**10×10^3**	**158×10^3**	**168×10^3**	**204×10^3**
Ferritin (24–336 ng/mL, for man)	**875**	**708**	**555**	**275**
CRP (0–0.5 mg/dL)	**0.25**	**0.24**	**0.23**	**0.52**
D-Dimer (0–500 ng/ml)	**6142**	**1415**	**440**	
INR (0.8–1, 24)	**1.02**	**1.12**	**1.14**	**0.99**
Fibrinogen (180–450 mg/dL)	354	225	235	180
LDH (135–225 u/L)	**1345**	**390**	**335**	**360**
Albumen (3.5–5.2 g/dL)	**3.6**	**3.82**	**3.7**	**4**
AST (0–40 u/L)	40	50	42	18
ALT (0–41 u/L)	40	135	85	32
ALP (40–130 u/L)	110	92	75	55
GGT (8–61 u/L)	160	210	225	185
Indirect billirubin	**1.45**	-	-	**0.28**
Total billirubin (0,3–1,2),	**1.80**	-	-	**0.45**
Prothrombin time (PT)	12	13	14	13
Activated partial thromboplastin time (a-PTT) (%)	42	25	22	23
Vitamin B12	300	-	-	350
Folic acid	7	-	-	>20
Thyroid-stimulating hormone (TSH)	1.5	-	-	-
Parathormone (PTH)	120	-	-	-
25-hydroxyvitamin D	10	-	-	-
Direct Coombs	**negatif**	-	-	-
Haptoglobin (0,3–2) g/L	**0.01**	**0.02**	-	**0.04**
Retuculocyte (0.52–3.53%) (Corrected)	**12**	**10**	-	**9.8**
K^+	3.5	3.8	4.2	4.5
Na^+	135	136	138	140
Ca^{+2}	7.6	8.2	8.7	9

Table 2. *Some serological analysis results of the patient*

Anti-HIV	0.25
HBsAg	3.8
HBeAg	0.910(0.9–1.1)
Anti HBS	167
Anti HCV	0.042
Brucella Aglütinasyon (Rose Bengal	negative
Brucella Aglütinasyon (With Coombs Anti Serum)	negative
SARS-COV-2	pozitive
Parvovirüs B19 (IgG, IgM)	negative
EBV	negative
VDRL-RPR	negative
Adenoviruses	negative
Anti-ds-DNA	9.5(0–25)
Anti-SSA	1.6(0–25)
Anti-SSB	3(0–25)
Anti-Scl-70	4(0–25)
Anti-JO-1	2(0–25)
Anti-CCP (cyclic citrullinated peptide)	9(0–20)
ANA (Antinuclear antibodies)	negative

man), d-dimer 6142(0–500 ng/ml), total bilirubin 1.80 mg/dL (0.3–1.2), indirect bilirubin 1.45 mg/dL, C-Reactive Protein (CRP) 0.25 mg/L (0–0,5), albumin 3.6 g/L (3,5–5,2), Aspartate Transaminase (AST) 40 u/L, Alanine Transaminase (ALT) 40 u/L, alkaline phosphatase (ALP) 110 (40–129 u/L), gamma-glutamyl transpeptidase (GGT) 160 (8–61 u/L), WBC 8100 cells/microL, neutrophil 51%, lymphocyte 35%, platelet count 10×103 (149–371×103 cells/microL), MCV 100 fL, INR 1, 02, pH: 7.43, SO_2: 96% HCO_3:21 mmol/L, $P0_2$: 62.4 mmHg and PCO_2: 31.5. The patient had no previous history of hematological disease. Diseases such as erythrocyte membrane disorders (such as hereditary spherocytosis), hemoglobinopathies (sickle cell or thalassemias), G6PD or (pyruvate kinase enzyme deficiencies) were excluded according to clinical, laboratory, and environmental blood analysis findings. Prothrombin time, activated partial thromboplastin time, and fibrinogen were within normal limits, and the direct Coombs test was negative. Peripheral blood smear showed spherocytic, polychromasic, and 8–10 fragmented erythrocytes (schistocytes) at each magnification (Figure 1). The brain, thorax, abdomen computerized tomography, and pulmonary angiography were unremarkable. No known organ transplantation or solid organ malignancy was detected in our patient. The patient's PLASMIC score was computed after the observation of a high level of schistocytes[3].

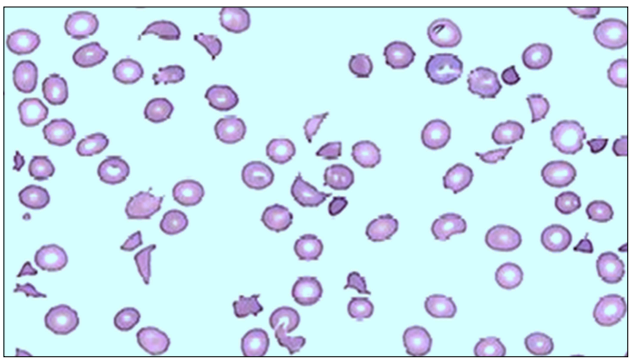

Figure 1. *Schistocytes in peripheral smear.*

Platelet count: 11×103 cells/microL (1 point if <30×103)

Haptoglobin: 0.03 g/L (1 point if <0.30–2 g/L)

[Other indicators of hemolysis Reticulocyte count (%) >2.5%; or Indirect bilirubin >2.0 mg/dL >34 mcmol/L],

If there is no active cancer: (1 point),

If there is no history of solid organ or hematopoietic stem cell transplant (1 point),

INR <1.5 (1 point),

Creatinine <2.0 mg/dL (1 point),

MCV <90 (0 point)

Our patient, who had a total of 6 points, was diagnosed with TTP. Serum samples were taken for ADAMTS13 and anti-ADAMTS13 IgG antibody levels, and the patient was started on 1 mg/kg prednisolone and 20 ml/kg/day (1600 ml) FFP. The next day, as the patient's values did not change, PLEX was planned, and he was referred to the hematology department. Plasmapheresis 50 ml/kg was administered there for three consecutive days, then five more times every other day. The prednisolone of the patient who was administered PLEX was also continued throughout this period and was tapered off. In addition, 0.5 mg Entacavir was started in the patient who was started on rituximab at 375 mg/m^2 per week (for four weeks) during this period and because he was HBsAg positive, HBeAg negative, and Anti-HBc positive (Table 2). On the 5th day of the treatment, PLT: 158×103, Hemoglobin: 9.1 g/dL, and LDH: 390 u/L levels, on the 30th day it was 204×103, 14.2, 362, respectively. The ADAMTS13 level obtained four months after the treatment was normal and was 73%. Currently using only 0.5 mg Entacavir, the patient was referred to the hematology clinic for eculizumab treatment planning.

Discussion

TMA refers to a pathological lesion seen on tissue biopsy. The presence of TMA is evident from clinical features such as MAHA and Thrombocytopenia with signs of organ damage. The median age in immune TTP is 40, which can be seen between the ages of 9–78. Its incidence is approximately 3 per million per year[6]. Although immune TTP is 30 times higher in adults, it can be seen in hereditary TTP. However, when TTP is suspected in the first pregnancy in an adult, hereditary TTP should not be forgotten[7]. In addition, TTP can be seen in autoimmune diseases such as SLE, and SLE is detected in 10% of immune TTP cases[8]. It has been suggested in previous studies that TTP cases in pregnant women are incorrectly confused with eclampsia and severe HELLP. Thrombotic thrombocytopenic purpura was more common in obese people, especially those with BMI >40 and women[6,9,10]. Thrombotic thrombocytopenic purpura may be immune-mediated due to autoantibodies against ADAMTS13 (anti-ADAMTS13 IgG antibodies) or inherited due to pathogenic variants in the ADAMTS13 gene. It is characterized by Thrombocytopenia, microangiopathic hemolytic anemia, and thrombocyte-rich thrombin in small vessels, sometimes causing organ damage. This condition causes Thrombocytopenia. Thrombotic thrombocytopenic purpura is an emergency that can be fatal if appropriate treatment is not started immediately. With proper treatment, more than 90 percent of patients survive[1]. Although TTP is a severe disease, some cases may present with nonspecific symptoms such as fatigue, dizziness, abdominal pain, nausea, and vomiting[11]. For this, a peripheral blood smear should be done first. If there is a schistocyte, it may be reasonable to calculate the PLASMIC score. PLASMIC score includes Platelet count <30.000/microL, Hemolysis (indirect bilirubin >2 mg/dL, reticulocyte count >2.5 percent or low levels of haptoglobin), absence of active malignancy, absence of bone marrow or solid organ transplantation, MCV <90 fL, INR <1.5, Creatinine <2.0 mg/dL. One point is given for each. If the total score is 4, the probability of TTP is low; if it is 5, it is likely TTP, and if it is 6–7 points, the likelihood of TTP is considered. Since the PLASMIC score is calculated after the presence of schistocytes is shown, it is not included in the scoring[12].

Calculated on a statistical probability, this scoring developed by Pavan Bendapudi and colleagues should be used cautiously. In TTP cases where patient mortality is high while waiting for ADAMTS13 level, using the PLASMIC score and starting treatment provides an advantage in terms of mortality. The 2020 systematic review confirmed the diagnostic accuracy of the PLASMIC score in patients with suspected TTP. The review identified 13 studies with a median TTP prevalence of 35 percent. A PLASMIC score of 5 or higher provided a sensitivity of 99 percent (95% CI 0.91–1.00) and a specificity of 57 percent (95% CI 0.41–0.72)[13]. ADAMTS13 level should not be considered alone for initiating or stopping treatment, as delay in diagnosis may increase the risk of mortality. However, the clinically diagnosed case can reveal the correct reason for the diagnosis[14]. Although ADAMTS13 activity in TTP is <10% in severe deficiency, ADAMTS13 may also be decreased in the presence of sepsis or systemic cancer. Intermediate values such as 10–59 may be seen in the presence of transfusion, sepsis, or systemic cancer. Normally, the activity should be >60%[15,16]. In immune TTP, deficient ADAMTS13 activity is caused by an inhibitör. A test for an inhibitor should be obtained for patients with severe deficiency. Inhibitor titer is determined from the number of serial dilutions of plasma after that inhibitor continues to inhibit ADAMTS13 activity. The titers are commonly presented in Bethesda units (the reciprocal of the dilution required to neutralize 50 % of the inhibitor). CM-TMA should be considered if diseases such as immune TTP, drug-related TTP, or SLE have been excluded in a patient as the test results will cause delay and threaten the patient's life. The ADAMTS13 level of our presented case was expected, and the patient was followed up in the hematology clinic for eculizumab treatment.

Conclusion

The peripheral smear should be performed in a patient with headache, dizziness, numbness in the arms, confusion, high fever, hemolytic anemia, Thrombocytopenia, high LDH, low haptoglobin, and increased reticulocytes. It is recommended to calculate the PLASMIC score to contribute to the diagnosis in a patient with a peripheral blood smear with schistocyte and normal levels of INR. Blood samples for ADAMTS13 and antibody levels should be obtained before starting treatment. Plasmapheresis should be applied if available. Steroids and rituximab should be given with PLEX. If PLEX is unavailable, the plasma infusion should be provided with steroids and rituximab. If CM-TMA is confirmed, eculizumab should be started.

Conflict of Interest Statement

All the authors declare no conflict of interest.

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