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BRIEF COMMUNICATION

NEPHROLOGY WILEY

COVID-19 associated thrombotic microangiopathy

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Abstract

A limited number of cases of thrombotic microangiopathy (TMA) have previously been reported in association with COVID-19. Our report describes two cases of TMA associated with COVID-19, one of which was successfully treated with eculizumab. The first case was a 23-month-old girl, and the second case was a 9-month-old boy. PCR tests for SARS-CoV-2 were positive in both cases, and laboratory results showed microangiopathic haemolytic anaemia, thrombocytopenia, and acute kidney injury. No known aetiology for TMA was found in either case. Stool tests for Shigatoxin-producing Escherichia coli were negative. Coagulation tests, ADAMTS13 activity, serum complement levels, and homocysteine levels were all within the normal range. No known genetic mutation was found, including mutations of complement, diacylglycerol kinase epsilon, and cobalamin C. In the first case, eculizumab was administered due to persistent haemolysis and prolonged anuria. In conclusion, TMA may be associated with COVID-19 infection. Treatment with eculizumab may be beneficial in selected patients because of the potential activation of the complement system.

KEYWORDS
COVID-19, eculizumab, HUS, thrombotic microangiopathy, TMA

1 | INTRODUCTION

Coronavirus disease 2019 (COVID-19), caused by the severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), has rapidly spread worldwide since its first description in December 2019.[1] Although the disease primarily affects the respiratory system, it is referred to as a multisystem disease due to its multiple manifestations in different organs. It is also associated with a prothrombotic state and an increased risk of thrombosis, including arterial and venous thrombosis.[2–4] In addition to macro thrombotic events, COVID-19 may also lead to microvascular injury and microvascular thrombosis, such as disseminated intravascular coagulation (DIC) and thrombotic microangiopathy (TMA).[5] It is considered that COVID-19-related TMA is due to endothelial injury, inflammatory immune response, and uncontrolled complement activation.[5–7] We report two cases of TMA associated with COVID-19, one of which was treated with eculizumab, an anti-C5 monoclonal antibody.

2 | CASE REPORTS

2.1 | Case 1

A previously healthy 23-month-old girl was admitted to the hospital with restlessness, vomiting, and diarrhoea characterized by watery

but not bloody stools that had persisted for 3 days. On admission, physical examination revealed her body weight of 11 kg (−1.17 SD) and height of 86 cm (−1.04 SD). She was anuric, oedematous, and severely hypertensive (blood pressure 125/95 mmHg, >95th percentile). She had a toxic appearance with fever and tachycardia. She had no respiratory disease with a normal chest radiograph, but SARS-CoV-2 PCR was positive.

The results of other laboratory tests showed severe anaemia, thrombocytopenia, and impaired kidney function (Table 1). Further anaemia studies revealed intravascular haemolysis, including elevated reticulocyte count and lactate dehydrogenase (LDH) level, low level of serum haptoglobin, and the presence of schistocytes in the peripheral smear. The patient had high levels of C-reactive protein and procalcitonin, while blood cultures showed no bacterial growth. The stool test for Shiga toxin producing Escherichia coli (STEC) was negative. ADAMTS-13 activity, coagulation tests, homocysteine, fibrinogen, and complement levels were all normal (Table 1). Renal ultrasound showed normal size of the kidneys but remarkable echogenicity of both kidneys and Doppler examination of the kidneys showed normal vascular flow patterns in the renal artery. Echocardiography showed no coronary artery dilatation or myocardial dysfunction.

TABLE 1 The laboratory findings at the time of the diagnosis of thrombotic microangiopathy.

Laboratory parameters	Case 1	Case 2
Haemoglobin (mmol/L)	4	4
Leucocyte count (cells × 10^9/L)	20.9	11.2
Lymphocyte count (cells × 10^9/L)	5.1	7.3
Platelet count (cells × 10^9/L)	24	38
Reticulocyte fraction	0.03	0.05
Direct Coombs testing	Negative	Negative
Haptoglobin (μmol/L)	0.12	0.12
Lactate dehydrogenase (ukat/L)	67.6	49.3
Urea (mmol/L)	35.4	17.9
Serum creatinine (μmol/L)	438	150
Alanine transaminase (ukat/L)	6.06	0.38
Aspartate aminotransferase (ukat/L)	2.61	1.34
C-reactive protein (mmol/L)	647.6	57
Procalcitonin (μg/L)	>100	0.75
Ferritin (nmol/L)	1.2	1.9
Complement 3 (μmol/L)	4.9	6
Complement 4 (μmol/L)	1.1	0.5
Prothrombin time (seconds)	14.5	14
Activated PTT (seconds)	25	23
D-dimer (mg/L)	7.4	38
Fibrinogen (g/L)	2.17	2.15
CK-MB (ukat/L)	1.11	0.51
Pro-BNP (pmol/L)	765.6	1693
ADAMTS-13 activity (%)	77	62
Homocysteine (μmol/L)	5	11

Note: Normal ranges of laboratory parameters: Haemoglobin 6.8–7.76 mmol/L, haptoglobin 3–20 μmol/L, Lactate dehydrogenase <2.83 ukat/L, C-reactive protein <47.6 mmol/L, procalcitonin <0.5 μg/L, ferritin 0.07–0.9 nmol/L, complement 3 (C3) 4.5–9 μmol/L, complement 4 (C4) 0.5–2 μmol/L, D-dimer <0.5 mg/L, fibrinogen 1.8–3.5 g/L, creatinine kinase myocardial band (CK-MB) <416 ukat/L, pro-brain natriuretic peptide (pro-BNP) <12 pmol/L, ADAMTS-13 activity >10%, homocysteine <15 μmol/L.

Due to anuria, volume overload, and uraemia haemodialysis treatment was initiated on admission. On the tenth day of her hospitalization, she was still anuric after two sessions of plasmapheresis in addition to supportive therapy. A renal biopsy could not be performed due to her unstable condition. On day 11, eculizumab was administered at a dose of 300 mg. Three days after eculizumab administration, the haemolytic episode ceased, no red blood cell transfusion was required, and kidney function improved. Haemodialysis was discontinued on day 20 (Figure 1A). Treatment with eculizumab was discontinued after nine months because the results of all genetic tests for complement mutations were negative, including complement factor H (CFH), complement factor I (CFI), complement factor B (CFB), thrombomodulin (THBD), plasminogen (PLG), C3, and CD46. The genetic tests were also negative for diacylglycerol kinase epsilon (DGKE), inverted formin 2 (INF2), and cobalamin C (CblC) mutations. At her last visit, 18 months after admission, she was not receiving any medications. Her kidney function was normal with a serum creatinine level of 25.6 μmol/L and no signs of hypertension or proteinuria.

2.2 | Case 2

A 9-month-old boy was admitted to the hospital complaining of vomiting, diarrhoea, and fever. The stool was not bloody. Physical examination revealed a body weight of 10.8 kg (1.27 SD) and a height of 80 cm (2.48 SD). He had a fever of 37.7°C. His blood pressure was normal. He had no symptoms of dehydration or volume loss. PCR testing for SARS-CoV-2 was positive. The respiratory system was unaffected, and chest radiograph was normal. On the third day of hospitalization, macroscopic haematuria, oedema, and hypertension were noted (blood pressure 110/80 mmHg, >95th percentile). Laboratory findings were suggestive of TMA with microangiopathic haemolytic anaemia (high serum LDH level, low serum haptoglobin level, and presence of schistocytes), thrombocytopenia, and acute kidney injury (Table 1). Laboratory tests for differential diagnosis of TMA revealed the following: Stool test for STEC was negative, ADAMTS-13 activity and serum levels of homocysteine and complement were all normal. Renal ultrasound showed increased echogenicity of both kidneys, and renal Doppler ultrasound was normal.

After two doses of plasmapheresis, an increase in haemoglobin and platelet count and a decrease in serum creatinine were noted without the need for dialysis. On the seventh day of his hospitalization, he was clinically recovered, and his laboratory values were normal. Fifteen days after hospitalization, he was normotensive, his serum creatinine was 24.7 μmol/L, and no proteinuria was detected

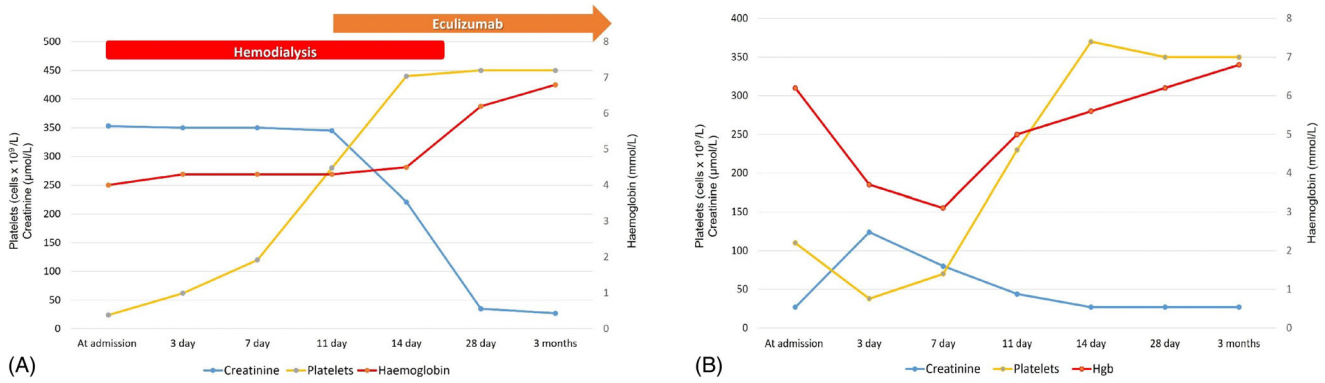

FIGURE 1 (A) Laboratory findings and acute kidney injury (AKI) course of the first case before and after eculizumab therapy. (B) Laboratory findings and acute kidney injury (AKI) course of the second case.

(Figure 1B). Genetic tests were negative for mutations in CFH, CFI, CFB, C3, CD46, THBD, PLG, INF2, DGKE, and CblC genes.

3 | DISCUSSION

A limited number of paediatric cases of TMA have been reported during COVID-19, of which only two were treated with eculizumab.[8–13] Our report also describes two children with TMA with no known aetiology other than confirmed COVID-19, one of whom was treated with eculizumab.

Both of our cases had clinical and laboratory findings consistent with TMA, including microangiopathic haemolytic anaemia, thrombocytopenia, and renal involvement during COVID-19. In the first case, inflammatory markers were too high, but there was no bacterial growth in blood cultures or no laboratory findings suggestive for DIC. No known aetiology for TMA was found in either case, including thrombotic thrombocytopenic purpura, typical haemolytic uremic syndrome (HUS) caused by STEC, complement independent atypical HUS (Cobalamin C defect-HUS, DGKE-HUS, INF2-HUS) or secondary HUS (no accompanying disease or medication use). In both cases, we could not explain TMA with complement-mediated atypical HUS (aHUS), since mutations were not detected in known genes, there was no family history, and no recurrence in the follow-up. However, it should not be forgotten that only 60%–70% of aHUS genes can be shown.[14] Taken together, we thought that TMA might be triggered by COVID-19 infection. This has been suggested by autopsy findings in patients with COVID-19 with diffuse microvascular thrombi like as in TMA.[15] The mechanism of endothelial injury in patients with COVID-19 involves activation of the alternative and lectin complement pathways, as confirmed by the deposition of membrane attack complex C5b-9 (MAC), C4d, and mannose-binding lectin-associated serine protease (MASP) 2 in the lung microvasculature of COVID 19 patients.[6] This observation was suggested by the detection of high plasma levels of soluble C5b-9 in COVID-19 patients.[16] Complement activation was also demonstrated by the detection of C5b-9 in peritubular capillaries, renal arterioles, and tubular basement membrane in COVID-19 patients.[17] Unfortunately, in our cases, we were unable to measure plasma levels of C5b-9 or perform renal biopsy, which is a limitation for providing conclusive evidence.

The observation of activation of complement system in these patients raised the question of whether eculizumab could be used as a treatment. Eculizumab is a monoclonal antibody against C5 that prevents the formation of C5b and thereby membrane attack complex on the surfaces. Eculizumab is effectively used to treat paroxysmal nocturnal haemoglobinuria and aHUS.[18] Short-term treatment with eculizumab has also been shown to be effective in patients with secondary TMA who had persistent symptoms despite treatment of the underlying cause.[19] It was reported from Italy that critically ill patients with COVID-19 patients with ARDS were successfully treated with eculizumab.[20] Laurence et al.[21] demonstrated the benefit of eculizumab treatment in three cases with COVID-19 proven complement dysfunction. Eculizumab was also used to treat cases with TMA due to COVID-19 infection, and it was highlighted that both haemoglobin levels and platelet counts increased rapidly, and serum creatinine levels decreased.[10,11,22]

In our first case, we administered eculizumab because of prolonged anuria and need for haemodialysis, and persistent haemolysis. We clearly observed that haemolysis ceased after the first dose of eculizumab, and serum creatinine decreased to baseline after the second dose. Although the patient had normal complement levels and no known mutations on complement genes, we observed the benefit of eculizumab, most likely indicating a contribution of complement activation to the pathophysiology of TMA in COVID-19. In the second case, we did not administer eculizumab due to clinical and laboratory improvement.

As a result, COVID-19 infection may be a trigger for TMA. Although we do not have evidence of complement system activation, the dramatic response to eculizumab suggests that the complement cascade is activated during COVID-19 infection. Further comprehensive studies are needed to clarify our findings.

CONFLICT OF INTEREST STATEMENT

The authors declare that there is no conflict of interest.

CONSENT FOR PUBLICATION

Written informed consent has been obtained from the parents.

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