Thrombotic microangiopathic haemolytic anaemia and antiphospholipid antibodies

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Objective: To analyse the clinical and laboratory features of patients with thrombotic microangiopathic haemolytic anaemia (TMHA) associated with antiphospholipid antibodies (aPL).

Methods: A computer assisted (PubMed) search of the literature was performed to identify all cases of TMHA associated with aPL from 1983 to December 2002.

Results: 46 patients (36 female) with a mean (SD) age of presentation of TMHA of 34 (15) years were reviewed. Twenty eight (61%) patients had primary antiphospholipid syndrome (APS). TMHA was the first clinical manifestation of APS in 26 (57%) patients. The clinical presentations were haemolytic-uremic syndrome (26%), catastrophic APS (23%), acute renal failure (15%), malignant hypertension (13%), thrombotic thrombocytopenic purpura (13%), and HELLP (haemolysis, elevated liver enzymes, and low platelet count in association with eclampsia) syndrome (4%). Lupus anticoagulant was detected in 86% of the episodes of TMHA, and positive anticardiolipin antibodies titres in 89%. Steroids were the most common treatment (69% of episodes), followed by plasma exchange (PE) (62%), anticoagulant or antithrombotic agents (48%), immunosuppressive agents (29%), and immunoglobulins (12%). Recovery occurred in only 10/29 (34%) episodes treated with steroids, and in 19/27 (70%) episodes treated with PE. Death occurred in 10/46 (22%) patients.

Conclusions: The results emphasise the need for systematic screening for aPL in all patients with clinical and laboratory features of TMHA. The existence of TMHA in association with an APS forces one to rule out the presence of the catastrophic variant of this syndrome. PE is indicated as a first line of treatment for all patients with TMHA associated with aPL.

Previously diagnosed APS may also develop TMHA. One of these reported cases was a patient with a 7 year history of SLE complicated by aPL who later developed TTP. Therefore, an association between aPL and the development of TMHA is clearly evident.

In this article we analyse the clinical and laboratory features of 46 patients—45 taken from published reports and one from our clinics—with TMHA associated with aPL, and support the hypothesis that TMHA might be a manifestation of the APS.

METHODS

A computer assisted (PubMed, National Library of Medicine, Bethesda, MD) search of the literature was performed to identify all cases of TMHA associated with aPL published in English, Spanish, and French from 1983 (when APS was first defined) to December 2002 (keywords: microangiopathic haemolytic anaemia, thrombotic microangiopathy, microangiopathic anaemia, thrombotic thrombocytopenic purpura, haemolytic-uremic syndrome, schistocytes, malignant hypertension, phospholipid, antiphospholipid, antithrombotic agents, anticoagulant, anticardiolipin antibodies, lupus anticoagulant, peptidase inhibitor, lupus inhibitor), and bibliographies of all articles.

Abbreviations: aPL, antiphospholipid antibodies; aCL, anticardiolipin antibodies; APS, antiphospholipid syndrome; FFP, fresh frozen plasma; HELLP, haemolysis, elevated liver enzymes, and low platelet count in association with eclampsia; HUS, haemolytic-uremic syndrome; LA, lupus anticoagulant; PE, plasma exchange; SLE, systemic lupus erythematosus; TMHA, thrombotic microangiopathic haemolytic anaemia; TTP, thrombotic thrombocytopenic purpura; vWF, von Willebrand factor.
were scanned for references not identified in the initial search. Only cases with well documented clinical summaries and relevant information were included. Data were summarised using a standardised data form, including sex, age, previous abortions or thrombotic events, immunological features, treatment, and evolution (web extra table W1, available at http://www.annrheumdis.com/supplemental).

For practical purposes, patients were included only if they had the following criteria: microangiopathic haemolytic anaemia, as indicated by erythrocyte fragmentation (schistocytes) on peripheral blood smears, with a negative Coombs’ test, thrombocytopenia, and presence of LA or positivity for aCL, or both. We categorised patients as having TTP if neurological dysfunction predominated, whereas patients with predominantly glomerular damage were diagnosed as the HUS.

To facilitate synthesis of these data we categorised patients in the following diagnostic categories:

- SLE, if they met four or more criteria of the American College of Rheumatology revised criteria for the classification of SLE; 1, 2
- “Lupus-like” syndrome if they met only two or three criteria
- “Primary” APS if they met criteria of the international consensus statement on preliminary classification criteria for definite APS syndrome; 18 and did not meet any of the above described criteria for SLE or lupus-like syndrome
- “Catastrophic” APS if they presented with an acute devastating APS with multiple organ involvement, as previously defined. 19

RESULTS
A total of 63 patients with TMHA associated with aPL were found in the literature search, but 18 of them (corresponding to six articles) 20–25 were not included because their clinical and immunological characteristics were not described. Because of this, 46 patients—45 from the literature 4, 5, 13, 14, 26–51 and one from our clinics (see appendix 1)—with 47 episodes of TMHA (one patient had two episodes of TMHA 26) were finally reviewed.

General characteristics
Table 1 shows the general clinical features of the complete series of patients. The patients comprised 36 (78%) women and 10 (22%) men with a mean (SD) age of 34 (15) years (range 7–73). Twenty eight (61%) patients had primary APS, 15 (33%) patients were categorised as having APS associated with defined SLE, 2 (4%) with lupus-like syndrome, and 1 (2%) with systemic sclerosis of paraneoplastic origin. During the follow up, the first diagnosis of primary APS was changed in three patients: one patient 26 developed a clinical picture of SLE 3 months later, another patient 27 developed a lupus-like syndrome 9 months later, and another 28 developed SLE 4 years later. In addition, one patient 29 had multicentric Castleman’s disease, and the necropsy of another patient disclosed a carcinoma of the uterus with pleural, bone, and hepatic metastasis. 30

Clinical presentation and precipitating factors
In 26 (57%) patients, TMHA was the first clinical manifestation of APS, whereas a total of seven (15%) patients had a previous history of major vascular occlusions. Deep venous thrombosis was reported as occurring in four (9%) patients, and in one this was accompanied by pulmonary embolism. Arterial occlusions occurred in two (4%) patients, arteriovenous fistula thrombosis in one, and haemodialysis vascular access and renal graft thrombosis in one patient. Spontaneous abortions or fetal death had occurred in 14 (39%) of the 36 female patients.

Table 2 shows the clinical presentation of 47 episodes of TMHA. HUS was the most common clinical presentation (26%), occurring in the postpartum period in three patients, and then catastrophic APS (23%), acute renal failure (15%), malignant hypertension (13%), TTP (13%), HELLP syndrome (4%), amaurosis fugax with microangiopathic haemolytic anaemia (2%), thrombocytopenia (2%), and thrombotic microangiopathy in renal allograft (2%).

In 21/46 (46%) patients, some precipitating factors contributed to the development of TMHA (table 3). This occurred during pregnancy in nine patients (fetal death in one patient) and in the postpartum period in six patients (one with acute enterocolitis). Major surgical procedures were evident as precipitating factors in three patients (one vascular surgery and two after renal transplantation), infection in two, and renal biopsy and oral contraceptive use (one case each).

Histopathological studies
Histopathological studies were performed in 32 patients (kidney biopsy in 27). The major finding was the presence of fibrin thrombi in glomerular capillaries in 18/24 (75%) patients, followed by double contours in glomerular capillary walls in 12/24 (50%), ischaemic glomeruli in 11/24 (46%), fibrin thrombi in arterioles in 11/24 (46%), and interlobular arteries in 8/24 (33%). In three cases the results of renal histopathological examination were reported only as “consistent with thrombotic microangiopathy”. In all cases, histological examination ruled out the presence of vasculitis.
Laboratory findings

Table 4 shows the laboratory pattern of the patients in this series. LA was detected in 31/36 (86%) episodes of TMHA in which this test was performed. The aCL titre was positive in 39/44 (89%) episodes of TMHA. This comprised all patients who tested positive for the IgG isotype of aCL, 50% of patients who were positive for the IgM isotype, and only one patient who was positive for the IgA isotype. Both LA and aCL positivity were detected in 22/33 (67%) patients. Antinuclear antibodies were positive in 15/29 (52%) patients. Anti-dsDNA antibodies were found in 7/24 (29%) patients, all of them with defined SLE.

Treatment and outcome

Table 5 shows the treatment of the 47 episodes of TMHA. For the statistical analysis, each episode of TMHA was considered separately, including those in the patients who had recurrences. Data on treatment were not available in five patients. Finally, 42 episodes of TMHA were analysed. Steroids (usually in high doses) were the most common treatment, used in 29/42 (69%) episodes. Plasma exchange (PE) was used in the treatment of 62% of episodes. Fresh frozen plasma (FFP) was given as replacement fluid in 13 (50%) episodes, together with normal saline in two, and with albumin in one; 5% albumin in two (8%) episodes; and in 12 episodes this information was not specifically reported. In four (10%) episodes, infusion of FFP without plasma removal was used as treatment. Twenty (48%) episodes of TMHA were treated with some form of anticoagulant or antithrombotic agent (heparin in nine, aspirin in eight, dipyridamole in four, and oral anticoagulant in three). Immunosuppressive agents were used in 12 (29%) cases (cyclophosphamide in 10, vincristine in two, and azathioprine in one). Intravenous immunoglobulins were used in five (12%). Intravenous prostaglandin E1 was used in one case. Most patients, however, received a combination of these treatments (web extra table W2, available at http://www.annrheumdis.com/supplemental).

When the presence or not of a single treatment is considered, recovery occurred in only 10/29 (34%) episodes treated with steroids (in all of them, steroids were used together with other treatments). In the seven cases in which steroids were the unique first treatment used, the clinical status and laboratory abnormalities worsened. Recovery occurred in 19 of the 26 (73%) episodes treated with PE. In nine patients this treatment was not the first treatment used, and its use correlated with a marked improvement of the laboratory abnormalities and patients’ clinical status. Specifically, one patient presenting with a clinical picture of TTP was initially treated with steroids. No change in her neurological status was seen and the platelet count decreased. PE was then used, and the patient’s neurological status improved and the platelet count rose. The same clinical course was seen in four additional patients, initially treated with steroids, heparin, and infusions of FFP and intravenous immunoglobulins. Indeed, one patient, presenting with malignant hypertension who had improved with PE, had a recurrence of thrombocytopenia upon withdrawal of PE.14 Another patient developed thrombocytopenia and worsening of her neurological status 3 weeks after the acute episode of catastrophic APS supervened. Intravenous immunoglobulin was added but her mental status continued to deteriorate. Monthly intravenous cyclophosphamide did not improve the clinical status and, finally, PE was restarted. Significant improvement was noted within 48 hours, and the patient responded to repeated PE over 3 years. Only in one case was heparin use directly related to an improvement in laboratory abnormalities and clinical status.9 In four cases this improvement appeared when heparin was used together with other treatments (in three of them PE). In the remaining four cases in which heparin use was not followed by a clinical improvement, PE was not used. Considering the four patients treated with infusions of FFP, in two of them no significant response was noted and the remaining two were discharged, but renal function remained impaired.

Of seven patients who presented with vascular occlusions before the development of an acute episode of TMHA, only three were receiving anticoagulant treatment (two of them, prolonged oral anticoagulant treatment, and the third patient, aspirin and heparin subcutaneously during pregnancy and recurrent spontaneous abortions).

Among the 46 patients, 10 (22%) finally died. Specifically, death occurred in three patients with clinical presentation of catastrophic APS, two with TTP, two with HUS, and
thrombocytopenia, malignant hypertension, and acute renal failure (one case each). The causes of death were catastrophic APS (three patients), infection (three patients), and myocardial infarction, massive cerebral haemorrhage, and massive haemorrhage after abdominal surgery (one case each). The remaining patient\(^2\) illustrates the difficulty of establishing the differential diagnosis between TTP and catastrophic APS. This patient presented with a clinical picture of SLE with diffuse proliferative glomerulonephritis and thrombotic microangiopathy. In the post-biopsy course, she developed shortness of breath with bilateral pleural effusions. Renal function continued to deteriorate and then generalised tonic-clonic seizure occurred. At this moment, thrombocytopenia and haemolytic anaemia with an increase in schistocytes was noted, and an LA was detected. Finally, arterial desaturation occurred with a synus bradycardia, which progressed to asystole and was followed by unsuccessful resuscitation. The postmortem examination of this patient showed extensive arteriolar and small arterial hyaline thrombi in multiple organs, including the myocardium, cerebral cortex, pancreas, kidney, and genitourinary tract. In some cases, the differential diagnosis between TTP and catastrophic APS remains difficult, and the patient described is an example of this problem (Appendix 1).

**DISCUSSION**

The term TMHA was first introduced to describe conditions in which localised or diffuse microvascular thrombosis occurs.\(^1\) TMHA encompasses a spectrum of disorders including TTP, HUS, malignant hypertension, postpartum renal failure, pre-eclampsia, and scleroderma renal crisis. The typical clinical picture may be complicated by thrombocytopenia, microangiopathic haemolytic anaemia, fever, neurological symptoms, and/or renal dysfunction, but not all these manifestations are required. Table 6 shows an attempt to establish a differential diagnosis of disorders that present with TMHA.

An intriguing question is whether aPL may have a role in the development of TMHA in patients with SLE. On the one hand, because aPL are present in up to 50% of patients with SLE,\(^3\) a positive aPL assay could be expected in a similar proportion of patients with SLE who develop TTP without necessarily implying an existing causal relationship. In a review of 28 patients with SLE and TMHA performed by Nesher et al;\(^4\) tests for LA or aCL were reported in eight patients, two patients had IgM aCL, and one had IgA antibodies on the solid phase enzyme linked immunosorbent aCL assays. Von Tempelhoff et al studied the incidence of acquired and/or inherited thrombophilia in 32 women with HELLP syndrome.\(^5\) Twenty two of them presented with aPL positivity; 17 with LA and 15 with aCL. They concluded that the aPL were the predominant thrombophilia defect, being present in 69% of patients with HELLP syndrome and thrombophilia defects. Most cases of HELLP syndrome are however aPL negative. Indeed, in our review, 28 (61%) patients were categorised as having a primary APS, and TMHA was the first manifestation of the APS in 26 (93%) of them. In the light of these data, we may conclude that in some patients (with or without SLE), aPL may play a part in the development of TMHA.

The aetiology of TMHA, especially in autoimmune diseases, is unclear. Systemic endothelial cell damage appears to be a central phenomenon in the pathogenesis of all TMHA syndromes. Direct evidence for this is the demonstration of apoptosis of microvascular endothelial cells in spleens removed from patients with TTP\(^6\) and the demonstration that plasma from patients with TTP or adult HUS can cause apoptosis of microvascular endothelial cells.\(^7\) Endothelial damage, regardless of its aetiology, may result in widespread release of unusually large von Willebrand factor (vWF) multimers. A plasma vWF-clearing protease (a metalloproteinase) has been postulated to decrease the size of large vWF multimers to their normal size in plasma after secretion. Deficiency of this protease has been reported in patients with acute idiopathic TTP,\(^8\) and an IgG autoantibody to the enzyme itself is responsible for its depletion. vWF-clearing protease deficiency may result in larger plasma vWF multimers that can cause platelet agglutination.\(^9\) However, these abnormalities may not be specific for idiopathic TTP. Other studies have shown that thrombotic episodes, especially arterial thrombosis, were more common in LA positive patients with low vWF-clearing protease activity.\(^10\) Moreover, Diez-Ewald et al found that vWF was significantly higher in patients with LA and thromboses and considered that the increased vWF was derived from endothelial cells damaged by LA.\(^11\) An abnormal vWF multimeric pattern was found in 71% of patients with multiple abortions and in 50% of those with strokes.\(^12\) These results suggest that decreased activity of vWF-clearing protease may be an additional risk factor for arterial thrombosis in patients with aPL. Trent et al described two patients with chronic relapsing TTP and aPL.\(^13\) The first was found to have unusually large vWF multimers in her plasma during TTP remission. Plasma of the second patient has not been analysed for the presence of unusually large vWF multimers. We could not include these two patients in our review because the aPL encountered were antiphosphatidylserine IgM and antiphosphatidylserine IgM respectively, with aCL and LA negativity. Mukai et al described a patient with arterial thromboses in APS.

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<th>Table 6 Differential diagnosis of thrombotic microangiopathic haemolytic anaemia</th>
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HUS, haemolytic-uraemic syndrome; APS, antiphospholipid syndrome; TTP, thrombotic thrombocytopenic purpura; CNS, central nervous system.
indicated for all patients with suspected TTP and HUS. Patients with malignant hypertension or HELLP syndrome (17% of the patients in our review) showed motor epileptic fits, and fever without any evidence of PE is the most important component of treatment and is TMHA, in general, may be the clinical presentation of an APS. In some patients, the diagnosis of TTP does not exclude the haemolytic anaemia in the absence of any evidence of catastrophic APS owing to the presence of microangiopathic infection or disseminated intravascular coagulation. The motor epileptic fits, and fever without any evidence of presentation. In one patient the diagnosis of TTP was based completely ruled out because a decrease in specific vWF-
molecule expression and monocyte adherence.70 However, other pathogenic mechanisms cannot be completely ruled out because a decrease in specific vWF-cleaving protease has been found in 89% of TTP patients but only in 13% of patients with HUS. In addition, Hashimoto et al were able to find from lupus prone mice a hybridoma clone producing an antibody able to induce thrombotic microangiopathy with some characteristics of TTP when injected into syngeneic mice.71

The clinical picture of TMHA, SLE, and APS may overlap and, if any of the two conditions coexist in the same patient, the diagnosis may be difficult at the time of initial presentation. In one patient the diagnosis of TTP was based on thrombocytopenia, microangiopathic haemolytic anaemia, motor epileptic fits, and fever without any evidence of infection or disseminated intravascular coagulation. The partial thromboplastin time was prolonged owing to the presence of LA. The authors excluded the diagnosis of catastrophic APS owing to the presence of microangiopathic haemolytic anaemia in the absence of any evidence of disseminated intravascular coagulation, but we consider that this patient might be categorised as having catastrophic APS. In some patients, the diagnosis of TTP does not exclude the diagnosis of catastrophic APS, but TTP specifically and TMHA, in general, may be the clinical presentation of an APS.

Patients treated with PE had a recovery rate of 73%. In fact, PE is the most important component of treatment and is indicated for all patients with suspected TTP and HUS. However, in contrast with TTP, where the infusion of healthy plasma in addition to the removal of the patient’s plasma, is very important for recovery, some published reports (exemplified in the patient reported here) suggest that in the case of catastrophic APS, removal of the plasma containing the damaging substance may be enough to establish remission without the need for the infusion of healthy plasma. Because plasma infusion is generally associated with more severe immediate adverse effects than infusion of 5% albumin, and because FFP may on occasion be associated with severe complications whereas albumin solution is almost free of side effects, it might be advisable to begin treatment of a catastrophic APS with PE using 5% albumin as a replacement fluid and, only when there is a lack of prompt response, consider the use of FFP. The value of additional treatments (especially antiplatelet drugs and steroids) is unknown. An appropriate randomised study to assess the role of steroids has not been carried out. Consequent on this analysis and review, we recommend, in cases of TMHA in association with APS, the use of PE as a first line of treatment. However, steroids should be still used in association with anti-coagulation, in order to limit or treat the massive cytokine release.

In conclusion, TMHA is a rare complication in patients with APS but it may be the first clinical manifestation of this syndrome. Furthermore, it may be part of the multiorgan failure syndrome seen in patients with catastrophic APS. This would support the need for systematic screening for LA and aCL in all patients with clinical and laboratory features of microangiopathic haemolytic anaemia. In addition, the existence of TMHA in an APS mandates the ruling out of the existence of the catastrophic variant of this syndrome. PE, perhaps using albumin as the initial replacement fluid, is the most important component of treatment and is indicated as first line of treatment for all patients with TMHA associated with aPL.

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APPENDIX 1

CASE REPORT
A 32 year old man was admitted with a 2 week history of confusion, headache, vomiting, abdominal pain, and fatigue. His previous medical and surgical history was unremarkable, and no drugs had been taken previously. On admission, the patient appeared confused with a bi-temporal headache. Blood pressure was 155/90 mm Hg and physical examination did not disclose any significant findings, except for a distended, non-tender abdomen. Bowel sounds were sluggish. Initial laboratory data included white blood cell count 9.6×10⁹/l with normal differential, haemoglobin 86 g/l, platelet count 48×10⁹/l, lactate dehydrogenase 1159 IU/l (normal range 250–450), haptoglobin 0.087 g/l (0.320–1.810), and serum creatinine 140 μmol/l (30–110). Both direct and indirect Coombs’ test were negative and peripheral blood smear showed many schistocytes. Urine analysis showed 2+ protein with mild haematuria (10 red blood cells/high power field). The prothrombin time and activated partial thromboplastin time were within the normal ranges, and normal d-dimer levels were found. Magnetic resonance imaging of the brain showed changes consistent with mild microvascular ischaemia in the cerebral white matter.

Soon after his admission, he developed episodes of an acute coronary syndrome (angina), and an electrocardiogram showed T wave inversion and Q waves on the inferior surface.
Creatine kinase and troponin T tests were normal. A myocardial-gamagrophy perfusion study showed mild anterolateral and inferior ischaemia. LA (determined following the guidelines of the Subcommittee for the Standardisation of Lupus Anticoagulants of the International Society of Thrombosis and Hemostasis) was present and aCL IgG, measured using standardised enzyme linked immunosorbsent assay (ELISA; Chesper Diagnostics, Chester, United Kingdom), was markedly raised at 65.9 IgG phospholipid (GPL) units (normal<15 GPL units). Antithrombin, protein C and protein S were normal, and factor VIII activity was 85% (normal 80–120%). Prothrombin time was 10.8 s (normal 10–14 s). Plasma levels of ADP and thromboxane A2 metabolites, and cryoglobulin levels were normal. Of altered mental status, microangiopathic haemolytic anaemia, thrombocytopenia, renal failure, and cardiac involvement, a presumptive diagnosis of catastrophic APS was made.

Treatment was started with PE using 5% albumin as replacement fluid (four sessions), intravenous prednisolone and coumadin adjusted to an anti-coagulant in systemic lupus erythematosus: a clinical and renal pathology perspective. Case report. J Rheumatol 1991;

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