The Epidemiology, Management, and Outcome of atypical Hemolytic Uremic Syndrome: Two Tertiary Hospital experiences

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Abstract

Objectives: Atypical Hemolytic-Uremic-Syndrome is a rare, life-threatening disease of chronic uncontrolled complement-pathway activation that leads to thrombotic-microangiopathy, along with **s**evere organ damage, including End-stage-kidney disease. This study aims to evaluate the epidemiology, management, and outcome of the atypical hemolytic uremic syndrome in a population from the Arabian Gulf.

Methods: This is a descriptive cohort study assessing all cases of atypical Hemolytic -Uremic Symdome **(**aHUS) from January 2008 to December 2019 based on clinical features, complement pathway assays, histopathological, and genetic testing among patients who had been diagnosed and followed up at two tertiary care centers in Oman.

Results: This study accrued 19 patients who fulfilled the inclusion criteria. Out of 19 patients, 57.9% were males, and 63% were adults, with a median age of 25 years (0.1-69). Fifteen (79%) presented in the acute phase of the disease, and 21% presented with clinical evidence of chronicity. A trigger factor was identified in 68.4% of patients. The triad of hemolytic anemia, acute kidney injuery(AKI) , and thrombocytopenia was present in all patients. A kidney biopsy was accepted by and performed in 74% of patients, 10 (71%) had aHUS in native kidneys and three in kidney grafts. Five patients showed features of acute thrombotic-microangiopathy while three had a combination of acute-on-chronic thrombotic-microangiopathy. Eleven patients underwent genetic analysis with 45.5% having a known pathogenic variant in atypical hemolytic uremic syndrome susceptibility genes. Plasma exchange followed by Eculizumab was the treatment method in 58% of cases, with complete renal recovery achieved in 50% of them. Finally, 21% died during the study period.

Conclusion: To date, this is the first report on the atypical hemolytic uremic syndrome in this particular population. It has a wide spectrum and multiple expressions which impose a significant challenge to diagnose and hence the commencement of the appropriate treatment at the onset. Eculizumab is the key to suppressing the complement activation; when afforded it should be the first-line therapy and delivered as early as possible. More studies with robust methodology are therefore warranted.

Keywords: Atypical hemolytic uremic syndrome, Epidemiology, outcome, Oman.

Introduction

Hemolytic uremic syndrome (HUS), is a sporadic disease characterized by hemolytic anemia, sudden or gradual onset of thrombocytopenia, acute kidney injury (AKI), and or extra-renal end-organ damage.[1,2] Furthermore, HUS is indicated by thrombotic microangiopathy (TMA) on histological examination.[3,4].Infection with Shiga-like toxin-producing Escherichia coli (STEC-HUS) has been documented to be responsible for more than 90% of cases, which carry a good prognosis.[5] On the other hand, patients with non-STEC-HUS tend to have poor prognosis indicators and it results in high mortality and morbidity rates in almost 50% of patients.[6] In this case, when untreated, patients remain at lifelong risk of kidney dysfunction, end-stage kidney disease (ESKD), extra-renal complications, and subsequent premature death. We have previously reported that HUS populations were mostly due to Shiga toxin-producing Escherichia Coli.[7] We showed that the HUS population was young, mostly male and only 25% have known medical comorbidities at the time of presentation. Also, the majority presented with acute kidney injury requiring dialysis, of which peritoneal dialysis was the mainstay of extant therapeutic modality. The duration of renal replacement therapy and recovery time was almost a month.[7]

Atypical Hemolytic Uremic Syndrome (aHUS) is an aggressive, lethal, and global disease, classified as a rare genetic disorder that stems from an inappropriate stimulation of the complement system.[8] It is termed "atypical" owing to the absence of an activating event similar to the conventional HUS, which activates on acquaintance with the Shiga-like toxin. The aHUS can also occur or manifest at any age, but this disease predominately affects children and young adults.[9]

The fundamental pathophysiology of aHUS is an uncontrolled stimulation of the complement pathway, which affects various vascular beds.[10] A quarter of patients succumb to death in the acute phase, and up to half progress to ESKD, which is exacerbated by dysregulation leading to glomerular endothelial cell damage.[11] Besides, up to 48% of patients have extrarenal manifestations with frequent neurologic and cardiovascular involvements. As a result, aHUS has been perceived to have feeble consequences. However, initial vigilance in aHUS identification, diagnosis, and instituting prompt treatment could lead to good prognostic indicators.[12]

Dysregulation of the alternative pathways of the complement system at various stages is a crucial part of the pathogenesis of aHUS.[13] The complement system, a part of the innate immune system, can be triggered through its three pathways: the classical pathway, the alternative pathway (AP), and the lectin pathway.[4,8,9,11] These three complement tracks congregate at the cleavage and activation of the fundamental complement C3. The initiation of the AP is securely controlled by numerous circulating and complement-cell-bound-complement regulatory proteins. An energetic equilibrium between complement activation and inactivation exists.[4,8,9,11] Genetic alterations play a role in such activation, and almost half of the aHUS-affected population has contributed to mutations in complement genes.[13-15] The loss-of-function mutations in regulators such as complement factor H (CFH), complement factor I (CFI), membrane cofactor protein (MCP), and thrombomodulin (THBD); and gain-of-function mutations in key complement components (complement component 3 (C3) and complement factor B (CFB)) prejudice the population to aHUS.[13-16] The genotype-phenotype relationships of aHUS have clinical implications for predicting kidney recovery and positive transplant results.[17-18] It is essential to report and validate these findings clinically and in the laboratory in as many populations as possible because the human genome has broad racial dissimilarities. Moreover, physical variances and physiological differences exist between people from various parts of the planet.[19] There are likely to be vast differences in features between people from different regions, ethnicities, or races.To date, there are limited studies done in Arabian Gulf countries. This study has embarked to evaluate the clinical and laboratory findings in the aHUS population seeking consultation from tertiary care centers in one such Arabian Gulf country, Oman.

Method

This is a descriptive cohort study evaluating all cases with the clinical diagnosis of aHUS between January,2008 and December ,2019, from two tertiary centers the Royal Hospital and Sultan Qaboos University Hospital in the urban setting of Oman. The present tertiary care centres often received referrals from all corners of the country since these two are the only equipped to provide comprehensive care for a spectrum of aHUS.

The HUS was defined by the triad of hemolytic anemia, thrombocytopenia, and acute kidney injury, aHUS were suspected in all patients with typical history but with negative stool cultures for Shiga-like toxin and normal ADAMTS13 activity above 10%. Both adult and pediatric populations were included. We excluded patients with the diagnosis of Shiga-toxin-related HUS and secondary TMA. The electronic medical records of the patient's

demographics, clinical characteristics, and laboratory tests including genetic analysis and histopathological tests were reviewed by using the hospital information system (HIS) at the time of initial presentation, three months, and at the last follow-up for renal outcome and death. All renal biopsies are submitted for light microscopic, immunofluorescence and electron microscopic examination. Light microscopy stains include hematoxylin and eosin, periodic acid schiff and Jones silver stain (all performed at 3 levels) in addition to a Miller elastic stain. The immunofluorescence panel includes IgA, IgG, IgM, C3, C1q, kappa and lambda. The molecular genetic testing was carried out through next-generation sequencing (NGS) gene panels in CPA, CAP, and/or CLIA-accredited diagnostic molecular genetics laboratories as per established protocol. Accordingly, the full gene sequencing was performed of the following genes: CFH, CFI, CD46, C3, CFB, and multiplex ligation-dependent probe amplification (MLPA) in CFH, CFHR1, and CFHR3 (Northern Molecular Genetics Service, New Castle upon Tyne, UK for four patients). Full gene sequencing, including 10 bp of flanking intronic sequences and deletion/duplication analysis, was included in the analyses of the following genes: ADAMTS13, C3, CD46, CFB, CFH, CFHR1, CFHR2, CFHR4, CFHR5, CFI, DGKE, PIGA, and THBD (Fulgent, Temple City, CA, USA for four patinets) and ADAMTS13, C3, CD46, CFB, CFH, CFHR1, CFHR2, CFHR3, CFHR5, CFI, DGKE, MMACHC, PIGA, PLG, THBD (Cento gene, Rostock, Germany for three patients).

Descriptive statistics were reported as frequencies and percentages for categorical variables. Continuous variables were described as median and ranges or as mean and standard deviations. P values less than 0.05 are considered statistically significant. All statistical calculations were performed with the STATA 13 software (version 13.0, Stata Corp, College Station, Texas 77845, USA).

This study was approved by the two local IRB for both participating centers ref no SQU-EC/153/18MREC1772,RH SRC 58/2018.

Results

There were 19 patients included in this study. Table 1 shows the patients' demographics. All patients were Omanis, with 11 (57.9%) males and 12 (63%) adults with a median age of 25 years (0.1-69). None of the diagnosed patients had a family history of aHUS. The majority (79%) of the patients presented in the acute phase of the disease. However, four (21% of the cases presented clinical evidence of chronicity. In 13 (68.4%) of the patients, a trigger factor was identified as shown in table 1. The frequency of the extra-renal manifestations is shown in table 2. The main CNS manifestation was a seizure, which occurred in six (31.6%) cases. Equally, encephalopathy occurred in six (31.6%) cases, followed by strokes in two (10%). Ocular manifestations like optic atrophy were also found in three (15.7) cases. The cardio-vascular manifestation was found in two cases, one had global hyperkinesia suggestive of dilated cardiomyopathy while the other case had mitral regurgitation. Elevated liver enzymes were present in five (26%). One patient at presentation was found to have liver cirrhosis with portal vein thrombosis. In addition, another patient presented with extensive deep vein thrombosis of the lower extremities. There were no cases of pancreatitis. Table 2 and 3 show the clinical and laboratory parameters. The triad of hemolytic anemia, AKI, and thrombocytopenia was present in most of the cases at the time of presentation. AKI with anemia was present in all cases, with a median creatinine level of 358 umol/l (45.2-1040) and a median Hb level of 8.0 g/dl (4.1-10.9). All patients had elevated LDH with a median of 570 u/l (245-299) and a low haptoglobin median of 28.0 u/l (10-150) , also thrombocytopenia was present in 18 (94.7%) with a median of 79fl (22-180). Severe thrombocytopenia of ≤ 30 was identified in three (15.7%) cases. The majority (73%) of the patients presented with high blood pressure with moderate to severe AKI. Ten (52%) of those who presented with severe AKI required dialysis. Proteinuria and microscopic hematuria were present in 18 (94.7%). with a median of 387.o5mglmmol(31.1-858) and nephrotic range proteinuria reported in 26%of cases. The complement activity assay for both C3 and C4 was reported in 18 (94.7%) cases. Six (32%) were found to have low C3 levels with a median value of 927mg/l (113-1370), and C4 activity with a median of 226.5 mg/l (150-423). Upon follow-up, persistently low C3, the level was found at 20% only.

Table 1: The various clinical data of the 19 patients with aHUS

Demographics Data	***N* (%)**
Age (year)	Median 25 (Range: 0.1-69)
Nationality	
• Omani	19 (100)
Gender	
• Male	11 (58)
• Female	08 (42)

Comorbid Disease	
• DM	02 (11)
• Hypertension	12 (63)
• Obesity	05 (26)
Clinical Presentation	15 (79)
• Acute	04 (21)
• Chronic	
Trigger Factors	13 (68)
▪ Infection	11 (85)
• Gastroenteritis	06 (55)
• Ricktesia	01 (9)
• Ecoli	01 (9)
• Upper Respiratory Tract Infection	02 (18)
• Mycoplmycoplasma	01 (9)
▪ Postpartum	02 (15)
▪ Post Kidney Transplant	03 (23)
▪ Antiphospholipid (APL)	01 (08)
▪ None	04 (31)

Table 2: The frequency of clinical and laboratory data on presentation.

On Presentation	
Clinical Data	**N (%)**
Seizure	**06 (31.6)**
Encephalopathy	**06 (31.6)**
Stroke	**02 (10.5)**
Ocular Manifestation	**03 (15.8)**
Cardiopulmonary	**02 (10.5)**
High Blood Pressure	**12 (63.1)**
Diffuse Vasculitis	**02 (10.5)**
AKI	**17 (89.4)**
CKD	**03 (15.8)**
LABORATORY DATA	
Anemia	**18 (94.7)**
Fragmented RBCS (Schistocyts)	**18 (94.7)**
Thrombocytopenia	**18 (94.7)**
Blood Film (Toxic Granulation)	**06 (31.6)**
Proteinuria	**18 (94.7)**
Microscopic Hematuria	**18 (94.7)**
ADAMTS13 Activity (>10%)	**19 (100)**

Results are expressed as numerical values, and percentages for categorical variables unless otherwise indicated.
***CKD** Chronic kidney Disease, **AKI** Acute Kidney Injury*

Table 3: The median of the laboratory parameters of a HUS at the time of presentation and 3 months follow-up.

Item	**On presentation**		***3 months follow-up***	
	Median	**Range**	**Median**	**Range**
Serological Evaluation				
Bilirubin (umol/L)	17	04-78	7	04-27
ALT (U/L)	18	05-253	12	05-189
ALP (U/L)	78	42-588	80	45-269
Albumin (g/L)	31	20-40	39	26-50
C-Reactive Protein (mg/L)	29.1	2.3-94.2	9	0.4-20
Urea (mmol/L)	28.8	4.2-49.9	7.1	2.3-16
Creatinine (umol/l)	358	45-1040	121	35-862
Urine PCR	387.05	31.1-8583.3	44.5	12.8-582
eGFR (mL/min/1.73)	10	02-37	41	04-100
Hemoglobin (g/dl)	8.0	4.1-10.9	11.6	7.3-13.7
Total Leucocytic Count (TLC)	7.6	4.5-14.8	5.8	2.4-12.8
Platelet (10^9/L)	79	22-180	237	64-346

Haptoglobin (u/l)	28	10-150	1230	390-2300
Lactate Dehydrogenase (LDH) (u/l)	570	245-2991	208	141-379
Prothrombin Time (PT) (sec)	11.1	9.6-16.4	11.1	10.2-17.4
Partial Thromboplastic Time (APTT) (sec)	33.8	25.8-48.0	35.55	30.2-46
Thrombin Time (TT) (sec)	17.8	13.4-22.4	16.1	11.6-20.8
Fibrinogen (g/l)	2.6	0.8-4.81	2.8	1.4-7.01
C3 (mg/I)	927	113-1370	960	240-1784
C4 (mg/I)	226.5	150-423	280	170-549

Results are expressed as numerical values, and medians (range) for continuous variables, unless otherwise indicated. ***eGFR*** *estimated glomerular filtration rate,* ***ALT*** *Alanine Aminotransferase,* ***ALP*** *Alkaline phosphatase,* ***C3*** *Complement component 3,* ***C4*** *Complement component 4.*

Classical pathway activities were performed in 11 (58%) cases and fell in the range of 34-115% (the normal range is 65-135%). Only 11% of classic pathway dysregulation was reported. Alternative pathway dysregulation was found in four (21%) with values in the range of 10-128%. Among patients with normal alternative pathway activity, one was already receiving treatment with Eculizumab. The test for s C5b-9 (MAC (membrane attack complex activity) was performed in 14 (73.68%) patients and six (31%) had elevated levels. Low Complement Factor H protein activity (*CFH/P*) was found in six (31%) cases with the range between 120-500ug when the normal value should fall between 284-528ug/ml. Autoantibodies (anti-factor H inhibitors) were found in five (26%), with the range of 3-420u/ml (Anti FH (IgG (auto) anti-factor H, Normal value <60 u/ml). ADAMTS13 activity was normal in all cases with normal activity defined as >10%.

A renal biopsy was performed in 14 cases (73%), and 10 patients underwent native renal biopsy for light microscopy, immunofluorescence, and electron microscopy. However, one patient was inadequately biopsied, features of acute thrombotic microangiopathy were reported in five cases, a combination of chronic acute thrombotic microangiopathy in three cases, and chronic thrombotic microangiopathy in one case. Of the 10 patients, three underwent kidney graft biopsy. One showed a recurrence at around one-year post-transplant with mild acute TMA, involving a few arterioles. There were no features to suggest rejection, and C4d was negative. Moreover, four patients without previous native kidney biopsies underwent post-transplant graft biopsies for impaired graft dysfunction which showed features of acute TMA and in two patients the C4d was positive.

Eleven patients underwent genetic testing (59%), five (45%) had a known pathogenic variant in aHUS susceptibility genes, and another four patients had variants of unknown significance (VUS) that were found alone in three of the five or in combinations with another pathogenic variant in two of them. Homozygous full gene deletions for the susceptibility genes *CFHR1* and *CFHR3* were identified in patients 1 and 6, whereas patient 2 was heterozygous for this deletion mutation making its role in the development of the HUS uncertain, as shown in Table 4. One patient (patient 4) carries a homozygous missense variant of unknown significance in the *CFHR3* gene (c.209T>C). Patient 3 is probably having PIGA-related NPH with aplastic anemia related to a somatic duplication in the PIGA gene. Patients 5 and 8 are heterozygous for pathogenic variants in the CD46 (c.175C>T, p.Arg59*) and CHF (p.Arg1206Cys) genes, respectively. Other identified VUSs include the CFH heterozygous intronic variant (c.965-6T>C) with an unknown splicing effect (patient 5), as well as the CFHR5 heterozygous variant (c.1561T>A, p.Leu521Ile) (patient 6), THBD heterozygous variant c.131C >T, p.Thr44IIe) (patient 7), and heterozygous variants in C3 (c.2901C >T, p.Leu967Leu) and *CFH* (c.3677C>A. p.Pro122Gln) genes identified inpatient nine, as shown in Table 4.

Table 4: The genetic analysis of patients with a HUS.

Patient No	Gene	Variant	Zygosity	Inheritance	Variant Classification And Predicted Role In Ahus Phenotype
1	*CFHR1,CFHR3*	1q31.1del	Homozygous	AD*/AR*	Pathogenic, susceptibility alleles in the homozygous state.
2	*CFHR1,CFHR3*	1q31.1del	Heterozygous	AD*/AR*	Pathogenic, susceptibility allele, uncertain increase in risk in the heterozygous state
3	*PIGA*	Whole gene duplication[a]	Hemizygous	Sporadic somatic mutations	Probably a somatic mutation that also explains the aplastic anaemia identified in this patient.
4	*CFHR3*	c.209T>C, p.IIe70Thr	Homozygous	AD*/AR*	VUS
5	*CD46*	c.175C>T, p.Arg59*	Heterozygous	AD*/AR*	Pathogenic, susceptibility allele
	CFH	c.965-6T>C	Heterozygous	AD/AR	VUS*
6	*CFHR1, CFHR3 CFHR5*	1q31.1del	Homozygous	AD*/AR*	Pathogenic, susceptibility allele in the homozygous state.
		c.1561T>A, p.Leu521Ile	Heterozygous	AD*	VUS*
7	*THBD*	c.131C >T, p.Thr44IIe	Heterozygous	AD*	VUS*
8	*CFH*	p.Arg1206Cys	Heterozygous	AD*/AR*	Pathogenic, susceptibility allele
9	*C3*	c.2901C >T, p.Leu967Leu	Heterozygous	AD*	VUS*
	CFH	c.3677C>A. p.Pro122Gln	Heterozygous	AD*/AR*	VUS*

AD: autosomal dominant, **AR: autosomal recessive, **VUS:** variant of unknown significance, a duplication segment appears to be larger and involves several genes on chromosome X but array comparative genomic hybridization was not done to define the exact size.*

Table 5 shows the pattern of treatment also the associated renal outcome and mortality at the end of three months, then at the last follow-up, with a median of 5 years (1-7). Plasma exchange followed by Eculizumab was the treatment strategy in 11(58%%).36% attained complete renal remission while the other 64% of the patients continued dialysis-dependent, of whom three underwent a kidney transplant. Plasma exchange alone was the treatment in one patient, and plasma infusion was the treatment choice in another, the remaining three patients were managed conservatively. Three pediatric patients received eculizumab as the first-line therapy. All three had complete hematological recovery and renal remission at the three-month follow-up. They sustained complete renal remission at the time of the last follow-up. Overall, there were four deaths (21%), including one patient at the three-month follow-up another death within the first year and the third after 3 years of follow-up, and the fourth death after 5 years of follow-up. All deaths were due to the disease process. Eculizumab was well tolerated despite a major event following an episode of meningitis in an adult patient, one year after Eculizumab treatment, despite a vaccination.

Table 5: The pattern of treatment and the associated renal outcome and mortality among the 19 cases of a HUS.

Treatment Options	**Total Patients N=19 N (%)**	**Initial outcomes (3 months)**				**Last Follow-up outcomes Median 5 (1-7) years**		
		Renal Remission N (%)	**CKD N (%)**	**ESRD/Dialysis N (%)**	**Death N (%)**	**Renal Remission N (%)**	**ESRD/Dialysis N (%)**	**Death N (%)**
Conservative Treatment	3 (16)	0	1 (33)	2 (67)	0	0	2 (67)	1 (33)
PE/PI	2 (10)	2 (100)	0	0	0	2 (100)	0	0
PE and Eculizumab	11 (58)	4 (36)	0	7 (64)	1 (9)	0	10[a] (100)	2 (20)
Eculizumab	3 (16)	3 (100)	0	0	0	03 (100)	0	0

Results are expressed as numerical values and percentages for categorical variables unless otherwise indicated.
PI *plasma infusion,* **PE** *plasma exchange,* **CKD** *Chronic kidney Disease,* **ESRD** *End-stage renal disease (requiring renal replacement therapy),* **NA** *not applicable*
[a]*Three patients went for kidney transplant*

Plasma exchange was given in the range of 5-10 sessions and Eculizumab was given as a second-line treatment after failure to respond to plasma exchange except in 3 pediatric patients. This was given as follows: 900 mg weekly for the first four weeks as induction, followed by maintenance doses of 1200 mg every fortnight. The three months follow-up laboratory response is shown in table 3.

Discussion

This is the first report of the clinical presentation and the genetic results of adult patients diagnosed with aHUS in Arab countries. Despite having a higher rate of consanguinity this study failed to find a familial trend in the diagnosed cases.[8] We had cases that presented with peculiar unreported clinical manifestations. Three patients had optic atrophy. This ocular manifestation is increasingly being reported in the literature but to date only limited to case reports.[20-21] One of the patients, a female, who had cardiac involvement continued to have volume overload and pulmonary edema. Her Echo reported severe mitral regurgitation (MR). This could stem from ischemia of the cordi tend. Another patient, a male, had a conjunctival hemorrhage, aHUS is usually a thrombotic disease, and hemorrhage could be due to thrombocytopenia or severe hypertension. However, his platelet was 22 10^9/L, which is not usual to be associated with spontaneous hemorrhage, hence was initially evaluated as a possible hemorrhagic fever and his serology for rickettsia was positive. One patient at the time of the diagnosis had clinical features of liver cirrhosis with portal vein thrombosis. This presentation is challenging as thrombocytopenia could be explained by liver cirrhosis. This patient received conservative treatment. Some of the patients had evidence of chronicity by history or renal biopsy at the time of the acute first presentation. This wide spectrum and multiple faces of the presentation impose a significant challenge to diagnose and hence commence the appropriate treatment at the onset. In the present cases, an infectious trigger was found in the majority.

In this study complement activation, and functional assay did not add a diagnostic value. However low C3 levels were found in 32% of patients at the time of presentation a finding similar to what is reported in the literature. However, a higher frequency of low C3 level was reported to be associated with C3 or CFB mutations.[10,14,15,22] Four patients with acute TMA post-kidney transplant did not have native kidney biopsies and the donor-specific antibodies (DSAs) were not performed which posed a diagnostic challenge to differentiate the TMA from antibody mediated rejection(AMR). Furthermore, two of the patients had C4d positive. Although half of the patients underwent molecular genetic testing, the yield was relatively high. The most frequent pathogenic variant was in *CFH*. This is similar to the result published by the Caucasian population.[2325]

The identification of the *CFHR1*, and *CFHR3* gene deletions in the homozygous state further confirms their role in the pathogenesis of a HUS.[26]The molecular genetic testing was done at different laboratories with variability in the number of genes and methodology used in these panels, which made the comparison of patient results not possible. Another issue is that segregation analysis was not performed to assess the pathogenicity of VUSs. However, segregation analysis for VUSs on a small number of family members will not be sufficient to support the pathogenicity of the variants given the fact the susceptibility genes have reduced penetrance, and further functional studies through protein expression assays will be needed to further establish the role of the variant in the subjects with VUSs. Given the severity of HUS in the six patients with VUS, establishing whether they are of pathogenic relevance is of clinical importance. Five out of the six variants were in *CFH* and the six were in C3. A higher frequency of the C3 variant was reported in the Asian population.[27] In this study, complete renal remission, at last, follow-up was achieved in 50% of the patient who received eculizumab. This rate is similar to what was reported by Fakhouri et. Al.[28] Among the seven who did not respond all were adults with clinical evidence of chronic disease and two were TMA in renal grafts. Of the seven patients who had no response to Eculizumab, three died. Three had genetic testing (patient numbers 1 and 8 in table 4). Patients, one and two had combined pathogenic mutation in *CFH* as well as Factor H autoantibodies. Patient number 1 was a female and she had a very aggressive course with extra-renal thrombosis, early recurrence of HUS after kidney transplant, and graft failure despite treatment with Eculizumab. On dialysis, she developed calcific uremic arteriolopathy and died five years after the diagnosis of aHUS. Patient two died after a massive ischemic stroke in the first year of diagnosis. While the third death was in a male (patient 8), who was diagnosed after a kidney transplant and died following a stroke 3 months after his transplantation. Eculizumab is more effective in the early treatment of aHUS.[11,22,29-30] Persistence of hemolysis or lack of improvement in renal function after 3–5 daily plasma exchanges has

been regarded as a criterion for uncontrolled TMA and is an indication for initiating eculizumab.[31] There are not many reported cases in the literature in which eculizumab was used as the first-line therapy of de novo aHUS in native kidneys. Ohanian et al. reported the use of eculizumab as first-line therapy in a severe case of aHUS complicated by CNS involvement.[32] In an open-label single-arm phase 2 trial, Fakhouri et al reported discontinuation of dialysis in 79% of patients treated with eculizumab as first-line treatment,[31] however, there is limited use of eculizumab as first-line treatment. This is like in our case we used as first-line in three cases with full recovery. This is due to limited access to the medication because of its high cost. The use of eculizumab in the treatment of secondary HUS is not yet established.[27] In our series, we treated a patient whose serology tested positive for rickettsia and had a worsening course of aHUS manifested severe enough to require intubation and ICU care despite treatment with doxycycline and plasmapheresis. After the first dose of eculizumab, dialysis was stopped and he was extubated. His genetic testing for compliments was negative.

Rickettsia infection is known to cause AKI and was reported to be associated with poor renal outcomes.[33] In this report, at postmortem examination, few patients revealed features of TMA. Albeit the use of eculizumab in the context of ongoing infection is worrisome and requires further investigation. Three patients received conservative treatment because they presented with established CKD and no features of active HUS. One of them died three years after the diagnosis due to a stroke. Screening of recurrent HUS post kidney transplant and after initiation of dialysis is essential particularly if treatment with eculizumab is discontinued. However, Stroke is one of the serious vascular manifestations of HUS that occur suddenly and is difficult to predict.[12] Hence, for patients with HUS who manifested with stroke or another serious cerebrovascular event one could make a case for a prolonged course of eculizumab treatment.

Limitations

As is often the case, retrospective studies tend to be marred with many factors that hamper their generation. First of all, segregation analysis was not performed to assess the pathogenicity of VUSs. Secondly, this study is limited by the fact that it only documents patients seeking consultation from tertiary care. Such populations are likely to have been those with more advanced pathology that warranted referral to the present tertiary care centers.

Conclusion

The atypical hemolytic syndrome in Oman as reported by others is a multiple-hit disorder. It has a wide spectrum and multiple faces of presentation which impose a significant challenge to diagnose and hence the commencement of appropriate treatment at the onset is paramount. The identification of the CFH1, and CFHR3 gene deletions in a homozygous state further confirms their role in the pathogenesis of aHUS. The VUSs were associated with aggressive disease and were likely to be pathogenic. Eculizumab is the key to suppressing complement activation and when afforded should be the first-line therapy and delivered as early as possible.

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