

First-Line, Early and Long-Term Eculizumab Therapy in Atypical Hemolytic Uremic Syndrome: A Case Series in Pediatric Patients

Selçuk Yüksel¹ · Havva Evrengül¹ · Z. Birsin Özçakar² · Tülay Becerir¹ · Nagihan Yalçın³ · Emine Korkmaz⁴ · Fatih Ozaltın⁴

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Abstract

Introduction Studies relating to first-line, early, and long-term eculizumab treatment and outcomes in children with atypical hemolytic uremic syndrome (aHUS) are scarce and unclear. The aim of this case-series study was to evaluate the outcomes of first-line, early, and long-term eculizumab treatment in our aHUS patients.

Materials and Methods We reviewed the data from four pediatric patients with aHUS who were treated with eculizumab. In three of them, eculizumab was used as a first-line therapy, and the follow-up period was ≥ 2 years in three patients.

Results Plasma exchange could not be performed in any patient. Plasma infusions were used only in Patient 1 (a 14-month-old boy) for 8 days without any response. Therefore, eculizumab was started on day 11 after admission. Patient 2 (a 16-month-old boy), Patient 3 (an 11-year-old girl), and Patient 4 (a 32-month-old girl) were treated with eculizumab as a first-line therapy, which was started 2–4 days after admission. The dosage of eculizumab was adjusted according to body weight. The hematologic parameters (the time frames were 3–17 days) and C_3 (the

time frames were 10–17 days) returned to normal in all patients after receipt of eculizumab. Although Patient 1 developed stage III chronic kidney disease, complete renal recovery occurred in Patients 2 and 4. Patient 3 also had reflux nephropathy with bilateral grade III vesicoureteral reflux and renal scars. Her creatinine clearance returned to the baseline value after receiving eculizumab. No complications related to eculizumab were observed in any patient during the follow-up period.

Conclusion Eculizumab can be successfully used as a first-line therapy in pediatric aHUS patients. We observed that the early initiation of eculizumab was associated with the complete recovery of renal function.

Key Points

Early and first-line initiation of eculizumab may be more effective for complete renal recovery in children with aHUS.

Ideally, treatment should begin within the first 4 days of presentation.

✉ Selçuk Yüksel
selcukyukse1.nephrology@gmail.com

¹ Department of Pediatric Nephrology, Pamukkale University School of Medicine, Kınıklı, Denizli, Turkey

² Department of Pediatric Nephrology, Ankara University School of Medicine, Ankara, Turkey

³ Department of Pathology, Pamukkale University School of Medicine, Denizli, Turkey

⁴ Department of Pediatric Nephrology and Nephrogenetics Laboratory, Hacettepe University School of Medicine, Ankara, Turkey

1 Introduction

Atypical hemolytic uremic syndrome (aHUS) is a systemic and life-threatening disease with poor prognosis that is defined by hemolytic anemia, thrombocytopenia, and acute kidney injury due to thrombotic microangiopathy [1]. The most important factors in the etiology of aHUS are disorders of complement regulation. In the past, aHUS without any treatment was associated with a fairly poor prognosis,

which was reported as 50 % of the patients progressing to end-stage renal disease and up to 25 % dying during the acute phase. Within the past 30 years, plasma therapy (plasma infusion [PI] and plasma exchange [PE]) has been used as a treatment modality in aHUS [2]. Although some patients with aHUS appear to respond to PI and PE, these approaches have frequently been found to be ineffective. Eculizumab, a humanized monoclonal antibody directed against complement C5, is an effective therapy in complement-related aHUS and could be beneficial in the long-term treatment of aHUS. Eculizumab therapy has frequently been used in cases of aHUS where the patients were dependent on, or resistant to, plasma therapy. Although a current international consensus report recommends the usage of eculizumab as a first-line therapy in pediatric aHUS patients, to date a very limited number of studies exist. These recommendations are not based on strong (large numbers) pediatric-based clinical research but, rather, on published data from adolescent and adult experience in particular [1–7].

In this report, we present four children with aHUS who were treated with eculizumab therapy. In three of them, eculizumab was used as a first-line therapy, and the follow-up period was ≥ 2 years in three patients.

2 Materials and Methods

The records of Turkish patients who were treated with eculizumab for the presumptive diagnosis of aHUS (i.e., microangiopathic hemolytic anemia, thrombocytopenia, and renal failure with hypocomplementemia, normal plasma ADAMTS13 activity, and without coexisting infectious or autoimmune diseases or Shiga toxin-producing *Escherichia coli* [STEC] infection) were reviewed. Creatinine clearance was calculated using Schwartz's formula. Proteinuria was assessed using 24-h urine protein excretion. For the genetic studies, DNA was extracted from peripheral blood following the standard phenol-chloroform protocol. The exons of *CFH* (NM_000186), *CFB* (NM_001710), *CFI* (NM_000204), *MCP* (NM_002389), *C3* (NM_000064), *CFHR 1-4* (NM_002113, NM_005666, NM_021023 and NM_001201550, respectively) and *DGKE* (NM_003647) genes, together with their adjacent intronic junctions, were analyzed by direct sequencing using BigDye v3.1 chemistry and an ABI3130 Genetic Analyzer (Applied Biosystems, Foster City, CA, USA) at the Hacettepe University Nephrogenetics Laboratory. Informed consent was obtained from each participant. The presence of Anti-Complement Factor H autoantibody was determined by CFH IgG ELISA Kit (Abnova), according to the manufacturer's recommendations, with a detection limit of 0.6 AU/m. ADAMTS13 activity was detected by

an ADAMTS-13 activity kit (Technozym) in accord with the manufacturer's recommendations with a detection limit of 0.2 % and an assay range of 0.3–105 %. Serum C3 and C4 concentrations were measured by the immunoturbidimetric method on Roche/Hitachi cobas c systems.

For verotoxin testing, stool samples or rectal swabs were inoculated onto both Sorbitol MacConkey (SMAC) and Cefixime and Tellurite-SMAC medium. Colonies were washed with sterile distilled water and placed into microcentrifuge tubes. For DNA isolation, tubes were incubated at 95 °C for 10 min. Samples were centrifuged at 10,000×g for 10 min and a supernatant was subsequently used as a source of DNA template. Reverse transcription polymerase chain reaction (RT-PCR) amplification was performed under the following conditions: initial DNA denaturation at 95 °C for 5 min, followed by 39 cycles of 95 °C for 1 min, 56 °C for 1 min, and 72 °C for 1 min. The final extension step was performed at 72 °C for 5 min. The amplified PCR products were run at 100 volts for 40 min for visualization by standard gel electrophoresis in a 2 % agarose gel [8, 9].

Eculizumab was given according to the manufacturer's recommendations: for body weights of 30 to <40 kg, 600 mg intravenously (IV) every 7 days for the first 2 weeks, followed by 900 mg IV for the third dose 7 days later, then 900 mg IV every 14 days. The instructions for body weights of 20 to <30 kg were to administer 600 mg IV every 7 days for the first 2 weeks, followed by 600 mg IV for the third dose 7 days later, then 600 mg IV every 14 days. For body weights of 10 to <20 kg, the manufacturer suggested 600 mg IV for the first dose, followed by 300 mg IV for the second dose 7 days later, then 300 mg IV every 14 days. Finally, for body weights of 5 to <10 kg, the recommended course of treatment is to administer 300 mg IV for the first dose, followed by 300 mg IV for the second dose 7 days later, then 300 mg IV every 21 days [7, 10].

The ethical committee of Pamukkale University approved the study protocol (approval number 60116787-020/35428).

3 Results

Since March 2012, we have encountered four patients with aHUS (age range: 14 months–11 years) (Table 1). PE could not be performed in any patient due to technical difficulties in our center. When the first patient (Patient 1, a 14-month-old boy) was admitted with aHUS in 2012, fresh frozen PIs were given for 8 days, and peritoneal dialysis had to commence. However, neither hematologic nor renal remission could be achieved. On day 11, eculizumab was started, and then urine output began. Hematologic parameters improved

Table 1 Clinical and laboratory findings of the patients at admission

| | Patient 1 (14-month-old boy) | Patient 2 (16-month-old boy) | Patient 3 (11-year-old girl) | Patient 4 (32-month-old girl) |
|--|---|--|----------------------------------|---|
| Findings at initial presentation | | | | |
| Body weight (kg)/height (cm) | 9/73 | 11/78 | 35/146 | 12/88 |
| Clinical presentation | Diarrhea (without blood), vomiting, edema | Diarrhea (with blood), vomiting, edema | Vomiting, headache, hypertension | Diarrhea (without blood), vomiting, edema |
| Urine output | Anuria (for 48 h) | Anuria (for 72 h) | 1.5 cc/kg/h | Anuria (for 72 h) |
| Hypertension (SBP/DBP) | No (90/70 mmHg) | Yes (120/80 mmHg) | Yes (140/100 mmHg) | No (90/70 mmHg) |
| Neurological signs | Seizure | No | No | No |
| Family history | No | No | No | No |
| Laboratory results on admission | | | | |
| Hemoglobin (g/dL) | 7.6 | 9 | 8 | 7.8 |
| Platelet count (per mm ³) | 69,000 | 190,000 | 84,000 | 60,000 |
| Urea/creatinine (mg/dL) | 70/2.2 | 163/5 | 73/3.2 | 76/3.3 |
| Lactate dehydrogenase (U/L) | 1604 | 1892 | 1091 | 2024 |
| Haptoglobin (mg/dL) | <10 (low) | 21 (low) | <10 (low) | <10 (low) |
| C ₃ (mg/dL) | 64 (low) | 67 (low) | 70 (low) | 68 (low) |
| ADAMTS-13 (%) | 106 | 105 | 144 | 107 |
| Plasma homocysteine level | Normal | Normal | Normal | Normal |
| STEC | Negative | Negative | Negative | Negative |
| Autoantibodies against factor H | Negative | Negative | Negative | Negative |
| Other cultures (blood, throat, urine, stool) | Negative | Negative | Negative | Negative |
| Kidney biopsy | TMA | TMA | TMA | Not performed |

Haptoglobin normal levels 30–200 mg/dL, C₃ normal levels 90–180 mg/dL

DPB diastolic blood pressure, *SBP* systolic blood pressure, *STEC* Shiga toxin producing *Escherichia coli*, *TMA* thrombotic microangiopathy

promptly within 2–3 days (Table 2). Although the serum creatinine level decreased gradually and peritoneal dialysis was stopped, creatinine clearance did not return to normal. When Patient 2 was admitted to our clinic in May 2013, the previous guidelines for the management of aHUS pointed out the need for plasma therapy to be started as early as possible (that is, within 24 h of the disease onset) [11, 12]. However, in 2012 Zuber et al. [13] suggested that eculizumab might be considered as a first-line therapy for a first episode of aHUS, particularly in children. After these observations were obtained from Patient 1 and the recommendations were extracted from the literature, we decided to use eculizumab as a first-line therapy without previous PIs in Patients 2, 3, and 4 (Table 2).

Eculizumab was started 4 days after admission in Patient 2 (a 16-month-old boy) and 2 days after admission in Patients 3 and 4 (an 11-year-old girl and a 32-month-old girl, respectively). The hematologic parameters and C₃ returned to normal in all patients (Table 2). Since Patients 2 and 4 were admitted with anuria, peritoneal dialysis was

also performed. The time interval between the initiation of eculizumab and the achievement of a normal urine output was 2 days in Patient 1, 12 days in Patient 2, and 14 days in Patient 4. Patients 2 and 4 completely recovered with eculizumab. Eculizumab therapy was stopped in Patient 2, at the request of his parents, because no mutation was found. Interestingly, Patient 3 was found to have an atrophic left kidney in an ultrasonographic evaluation upon her first admission. We learned from her past medical history that she had suffered from recurrent febrile urinary tract infections. There were multiple scars bilaterally and an atrophic left kidney in DMSA (dimercaptosuccinic acid) scintigraphy. Voiding cystourethrography showed bilateral grade III vesicoureteral reflux. A biopsy was performed from the normal-sized right kidney, which revealed thrombotic microangiopathy (Table 2). After eculizumab therapy, her hematologic parameters returned to normal levels. Her serum creatinine level also returned to a basal level of 0.9 mg/dL, which was detected during a routine medical checkup 6 months previously. While eculizumab

Table 2 Treatment modalities and follow-up of the patients

| Treatment | Patient 1 (14-month-old boy) | Patient 2 (16-month-old boy) | Patient 3 (11-year-old girl) | Patient 4 (32-month-old girl) |
|--|--|--|---|--|
| Duration of FFP infusion and response | 8 days No response | Not used | Not used | Not used |
| PE | No | No | No | No |
| RRT | Peritoneal dialysis | Peritoneal dialysis | No | Peritoneal dialysis |
| Duration of RRT (days) | 11 | 8 | No | 20 |
| Complication of RRT | No | Peritonitis (<i>Streptococcus parasanguinis</i>) | No | No |
| Eculizumab | | | | |
| Time between admission and first eculizumab (days) | 11 | 4 | 2 | 2 |
| Dosage | 300 mg IV for the first dose, followed by 300 mg IV for the second dose 7 days later, then 300 mg IV every 21 days | 600 mg IV for the first dose, followed by 300 mg IV for the second dose 7 days later, then 300 mg IV every 14 days | 600 mg IV every 7 days for the first 2 weeks, followed by 900 mg IV for the third dose 7 days later, then 900 mg IV every 14 days | 600 mg IV for the first dose, followed by 300 mg IV for the second dose 7 days later, then 300 mg IV every 14 days |
| Time between first eculizumab and hematologic recovery (days) | 3 | 12 | 4 | 17 |
| Time between first eculizumab and C ₃ recovery (days) | 10 | 15 | 17 | 17 |
| Time between first eculizumab and normal urine output (days) | 2 | 5 | – | 10 |
| Adverse effect | No | No | No | No |
| Ongoing eculizimab therapy | Yes | No | Yes | Yes |
| Follow up period (months) | 41 | 34 | 24 | 16 |
| Relapse | No | No | No | No |

FFP fresh frozen plasma, PE plasma exchange, RRT renal replacement therapy

Table 3 The last physical and laboratory evaluations and genetic results

| | Patient 1 (14-month-old boy) | Patient 2 (16-month-old boy) | Patient 3 (11-year-old girl) | Patient 4 (32-month-old girl) |
|---|---|---------------------------------|--|---|
| Last physical examination | | | | |
| Age at the last follow-up visit | 55 months | 50 months | 13 years | 48 months |
| Height velocity (cm/year) | 6 | 7 | 11 | 8 |
| Hypertension (treatment) | No (enalapril) | No | No (enalapril) | No |
| Last laboratory tests | | | | |
| Hemoglobin (g/dL) | 10.6 | 11.1 | 12.8 | 13.3 |
| Platelet count (/mm ³) | 178,000 | 304,000 | 213,000 | 250,000 |
| Urea/creatinine (mg/dL) | 85/1.2 | 31/0.4 | 35/0.94 | 20/0.44 |
| Lactate dehydrogenase (U/L) | 233 | 286 | 198 | 260 |
| C ₃ (mg/dL) | 115 | 89 | 92 | 98 |
| Proteinuria (mg/m ² /h) | 20 | 2 | 30 | 2.5 |
| Creatinine clearance (mL/min/1.73 m ²) | 40 | 118 | 50 | 123 |
| Comorbidities | – | – | Bilaterally reflux nephropathy | – |
| Genetic screening | Heterozygous splice mutation in <i>CFH</i> gene (c.3133+1G>A) | No mutation | Two heterozygous variations in <i>C3</i> gene (c.3671G>A and c.4100T>C) Two heterozygous variations in <i>MCP</i> gene (c.285T>C and c.286+18_27delT) | A heterozygous variation in <i>MCP</i> gene (c.1148C>T) |

Creatinine clearance was calculated using Schwartz's formula

treatment was continuing, vesicoureteral reflux was treated with a bilateral subureteric injection in month 6 of the follow-up period.

Kidney biopsies were performed in Patients 1, 2, and 3 and revealed thrombotic microangiopathy. The reasons for biopsy in all patients were to confirm the diagnosis before starting eculizumab. In addition to Patient 1, who did not respond to plasma therapy, Patient 3 had an atrophic left kidney. We observed that glomerular changes (focal reduplication of the glomerular capillary basement membrane, thickening of the capillary walls) and interstitial changes (fibrosis and chronic inflammation) were predominant in Patient 1; glomerular changes (narrowing of capillary lumens, thickening of the capillary walls, congestion, thrombosis, necrosis) and vascular changes (arteriolar thrombosis, intimal thickness, occlusion) were predominant in Patient 2; and glomerular changes (thickening of the capillary walls, narrowing of the capillary lumens, congestion, mesangiolysis) and interstitial changes (acute inflammation and edema) were predominant in Patient 3.

All patients were vaccinated for *Neisseria meningitidis* before the initiation of eculizumab therapy and received antibacterial prophylaxis (macrolides or ceftriaxone) for 2 weeks. The follow-up periods of the patients without complications and relapses were 41, 34, 24, and 16 months, respectively. Eculizumab therapies are ongoing in Patients 1, 2, and 4, but Patient 2 has not been using the therapy for 30 months.

Mutation analyses were completed in all patients. A heterozygous splice site mutation in *CFH* in Patient 1 and four possibly relevant heterozygous variations in *C3* and *MCP* in Patient 3 were identified. Of these, two were already in the Single Nucleotide Polymorphism Database (dbSNP) (rs11569534 and rs11569541) with unknown significance. Except for Africans, these variations are rare in most populations, including Turks (i.e., none of the 2460 alleles were in the Turkish Exome Database). In Patient 3, two variations in *MCP* were identified. Of them, the c.285T>C variation was predicted to disrupt the splicing event according to Human Splicing Finder (HSF) software. Similarly, using the same software, the c.286+18_27delT variation is predicted to potentially alter the splicing event. However, as we have no data regarding CD46 surface expression analysis, their pathological significance remains questionable. In Patient 4, a heterozygous variation (c.1148C>T) was identified in *MCP*. No mutation was found in Patient 2. Genetic results are detailed in Table 3.

4 Discussion

In this report, we present four patients with aHUS who were treated with eculizumab. We used eculizumab in three patients as a first-line therapy in an early period of

their treatment. Two of them (Patients 2 and 4) have fully recovered. Patient 3 had chronic kidney disease due to reflux nephropathy, and we observed that her creatinine level returned to the baseline level with eculizumab therapy. In Patient 1, eculizumab was started on day 11 after admission. Currently, he has stage III chronic kidney disease. Our data indicate that three of the four patients improved with early initiation of eculizumab; however, in Patient 1, a partial response could be related to the late onset of therapy.

It is well known that disorders of the complement system, particularly the alternative pathway, are the most important underlying causes of aHUS [14, 15]. Autoantibodies against complement regulatory system proteins and defects in complement genes have been reported in cases of aHUS [1]. For whatever reason, the alternative complement system pathway is continually activated, and this process leads to the formation of the membrane attack complex. For this reason, theoretically, any defective complement protein or antibodies against complement proteins can be removed and replaced by PE. In addition, PIs can replace the defective complement components [16]. Thereby, according to the 2009 guideline suggested by the European Pediatric Study Group for HUS and some experts, the first-line treatment of aHUS should be plasma therapy (PE or PI) and should be started as early as possible [1, 11, 12, 17]. If PE is not performed within 24 h of first presentation, PI should be performed. If the patient has volume overload and/or severe hypertension, PI should be avoided. In addition, technical difficulties and equipment problems may limit the availability of PE [1, 18]. Another problem related to PE may be some unpleasant complications, such as thrombosis and infections, especially if used over the long term [1]. Moreover, in many countries, there are a limited number of technically trained centers that can perform PE, particularly in small children.

In the past decade, many patients with aHUS have also been treated with eculizumab. Although there are many reports about the use of eculizumab in medical literature, the vast majority of them involved patients who did not respond to the traditional approach of PE or PI [14, 19–27]. On the other hand, some authors recommended the early use of eculizumab for patients with aHUS who have a positive family history, recurrent HUS, or hypocomplementemia at presentation [27]. In our study, all of the patients were admitted to our clinic with hypocomplementemia before September 2014. We had to use eculizumab as a first-line therapy due to technical problems associated with PE. We observed that the usage of eculizumab in children with aHUS was efficient and safe for a relatively long period. Until recently, a very limited number of case reports of treatment with eculizumab as a first-line therapy have been published [5]. Finally, in 2016, in terms of the management of aHUS in children, a new

international consensus report has proposed eculizumab as first-line and early treatment (when possible, within 24–48 h of onset) [7]. The authors of the consensus re-evaluated four prospective, open-label, single-arm, non-randomized, multinational trials of the efficacy of eculizumab in aHUS patients [28–31]. The results of the first two trials were obtained from published data [28, 29], the others from preliminary data (abstracts) [30, 31]. However, three of four prospective trials consisted of adolescents (age >12 years) and adults [28, 29, 31]. One of them had addressed children with aHUS (22 patients, age \geq 1 month to <18 years) [30]. The recommendations of the 2016 consensus were supported with case reports of eculizumab to treat aHUS involving native kidneys. Among approximately 35 case reports of the patients who received eculizumab, 19 patients were children (aged 11 days to 11 years) and only five patients received first-line eculizumab. Thus, the 2016 consensus recommendations are intriguing, but these recommendations are not based on strong (large numbers) pediatric-based clinical research, particularly in terms of infants with aHUS [7]. Three of four cases in our study are infants and we know that published data related to this group are very limited. As a result, our findings can support and contribute to the recommendation of the 2016 consensus which reported that “first-line and early eculizumab is effective and safe in children with aHUS for renal recovery.”

We know that glomerular, vascular, and tubulo-interstitial changes vary according to the severity and the duration of the disease in patients with aHUS. Habib et al. [32] mainly distinguished three pathologic categories: cortical necrosis, predominantly glomerular involvement, and predominantly arterial involvement. The prognosis was the best in the patients with predominantly glomerular involvement; it was the worst in the patients with predominantly arterial involvement. In addition, many researchers have confirmed that vascular lesions (arteriolar and arterial) are a harbinger of a poor prognosis [33]. Although Patient 2 had severe vascular lesions, he clinically recovered completely with early eculizumab treatment. Patient 1 experienced the reduplication of the glomerular capillary basement membrane, which was defined as a chronicity sign, and he did not recover completely with eculizumab. These findings may indicate that early eculizumab treatment may correct the conditions known to be associated with poor prognosis in aHUS patients.

Meningococcal meningitis is the most feared side effect in patients treated with eculizumab. In addition, some other side effects, such as hematological abnormalities, nausea, vomiting, diarrhea, headache, hypertension, and recurrent upper respiratory tract infections, have been reported [16]. None of them were observed in our patients.

In Patient 1, c.3133+1G>A variation in *CFH* was at the consensus site. In silico analysis using HSF software

predicted a “Broken WT Donor Site” [34]. This variation was not detected in 2460 alleles obtained from the Turkish Exome Database. Therefore, it was considered a ‘disease-causing’ variation. We identified heterozygous c.3671G>A (p.G1224D) (rs11569534) and c.4100T>C (p.I1367T) (rs11569541) variations in *C3*, as well as heterozygous c.285T>C (p.Y95Y) and c.286+18_27delT variations in *MCP* in Patient 3. Of them, the c.3671G>A (p.G1224D) variation seems to be harmless, as predicted by SIFT and PolyPhen software (scores 0.33 and 0.04, respectively). Its minor allele frequency is 1 %; however, in certain populations such as Africans, it reaches as high as 3 %. The c.4100T>C (p.I1367T) variation is extremely rare (minor allele frequency [MAF] <0.01) in most populations, except for Africans, and is predicted to be deleterious according to SIFT software [35]. As our patients were Turkish, we looked at the frequency of both variations in the Turkish Exome Database composed of 2460 alleles and found neither of them in any of these alleles. Therefore, these variations were considered to be rare in the Turkish population as well. Although synonymous, the c.285T>C variation in *MCP* may disrupt the splicing event as predicted by HSF software since this position is very close to the splice site [34]. Similarly, using the same software, the c.286+18_27delT variation is predicted to potentially alter the splicing event. Allele frequency of the former in the Turkish population was determined as 2/2460 alleles (0.08 %) using the Turkish Exome Database. However, this could not be determined for the latter as this region contains poliT, which most likely causes reading errors in the next generation sequencing. We acknowledge that we cannot conclude their pathogenicity without determining CD46 surface expression. Combined genetic abnormalities in aHUS have been identified in patients with aHUS [36]. Thus, we speculate that the clinical phenotype might be related to the joint effects of these combined variations. To prove this, functional studies are needed. The c.1148C>T (p.T283I) that was identified in Patient 4 was previously described by Provaznikova et al. [37].

In our country, eculizumab is fully funded by the government. Therefore, the commission that is appointed by the Ministry of Health decides whether or not to approve the usage of eculizumab in a patient with aHUS. Moreover, if eculizumab is going to be continued, this procedure has to be repeated every 3 months. Eculizumab is a very expensive drug for Turkey, as in many countries of the world. The cost of eculizumab in a 300-mg vial is €4600. Due to this price, the annual costs of the treatment per patient are about €83,000 in Patient 1, €360,000 in Patient 2, and €120,000 in Patient 4, not including induction therapy at the onset of the disease.

5 Conclusion

Eculizumab can be successfully used as a first-line therapy in aHUS patients. Moreover, it seems that the early initiation of eculizumab was associated with the complete recovery of renal function. However, the cost of eculizumab therapy is an important limiting factor in the treatment of these patients. In addition, the questions of who should be treated and when should treatment be started and ceased still merit answers. Further studies are needed to better understand whether early eculizumab treatment could provide rescue therapy for patients with aHUS.

Acknowledgments The authors would like to acknowledge Professor Dr Aynur Karadenizli (Kocaeli University Faculty of Medicine, Department of Medical Microbiology) for verotoxin testing.

Compliance with Ethical Standards

Conflict of interest Dr. Selçuk Yüksel, Dr. Havva Evrengül, Dr. Z. Birsin Özçakar, Dr. Tülay Becerir, Dr. Nagihan Yalçın, Dr. Emine Korkmaz, and Dr. Fatih Özaltın have no financial disclosures or conflicts of interest that are directly relevant to the content of this article.

The data for this article was obtained from patient records. Informed consent for genetic testing and for publication of their child’s clinical information was obtained from the parents.

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