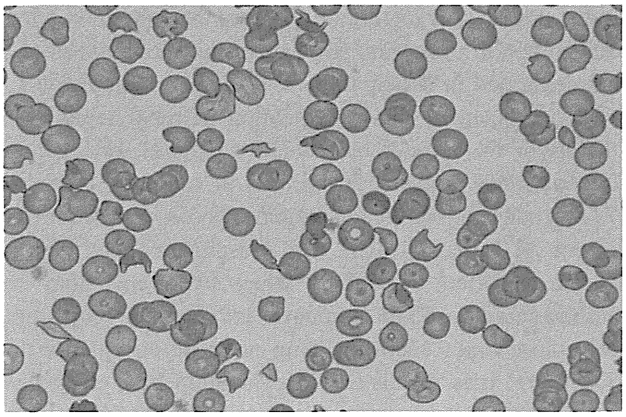

Fig. 4 Poikilocytes in a patient with HUS (×400)

Supplementary articles

a. UpToDate: Clinical manifestations and diagnosis of Shiga toxin associated (typical) hemolytic uremic syndrome in children. (Accessed on December 1, 2012)
b. Frank C, et al. Epidemic profile of Shiga-Toxin-Producing Escherichia coli O104:H4 outbreak in Germany. N Engl J Med 2011;365:1771–1780.
c. Loos S, et al. An outbreak of Shiga toxin-producing Escherichia coli O104:H4 hemolytic uremic syndrome in Germany: Presentation and short-term outcome in children. Clin Infect Dis 2012;55:753–759.
d. Trachtman H, et al. Renal and neurological involvement in typical Shiga toxin-associated HUS. Nat Rev. Nephrol;8:658–669.
e. Uemura O, et al. Age, gender, and body length effects on reference serum creatinine levels determined by an enzymatic method in Japanese children: a multicenter study. Clin Exp Nephrol 2011;15:694–699.
f. Kidney Disease Improving Global Outcomes (KDIGO) Acute Kidney Injury Work Group. KDIGO Clinical Practice Guideline for Acute Kidney Injury. Kidney Int Suppl. 2012;2:1–138.

2.2 Assessment of acute kidney injury (AKI)

AKI is a severe complication in HUS patients. 50 % of HUS patients manifest oliguria or anuria. Patients with oliguria or anuria require renal replacement therapy (acute blood purification). [Grade of Recommendation: Not Graded]

Risk factors of oliguria, anuria or hemodialysis are: dehydration, lack of isotonic fluid administration before development of HUS, hyponatremia (≤130 mEq/L), increase of serum ALT (≥70 IU/L) and infection by STEC O157:H7. [Grade of Recommendation: Not Graded]

When serum creatinine level is two times higher than age-sex standard, we suggest transferring the patient to a hospital where renal replacement therapy (acute blood purification) can be performed. [Grade of Recommendation: B]

Comments

1. AKI in HUS: epidemiology and pathophysiology

Over half of the patients with HUS manifested AKI. A Japanese survey reported that 47 % of the 132 patients with HUS manifested oliguria or anuria, and 27 % of the 132 patients had received renal replacement therapy [1]. In

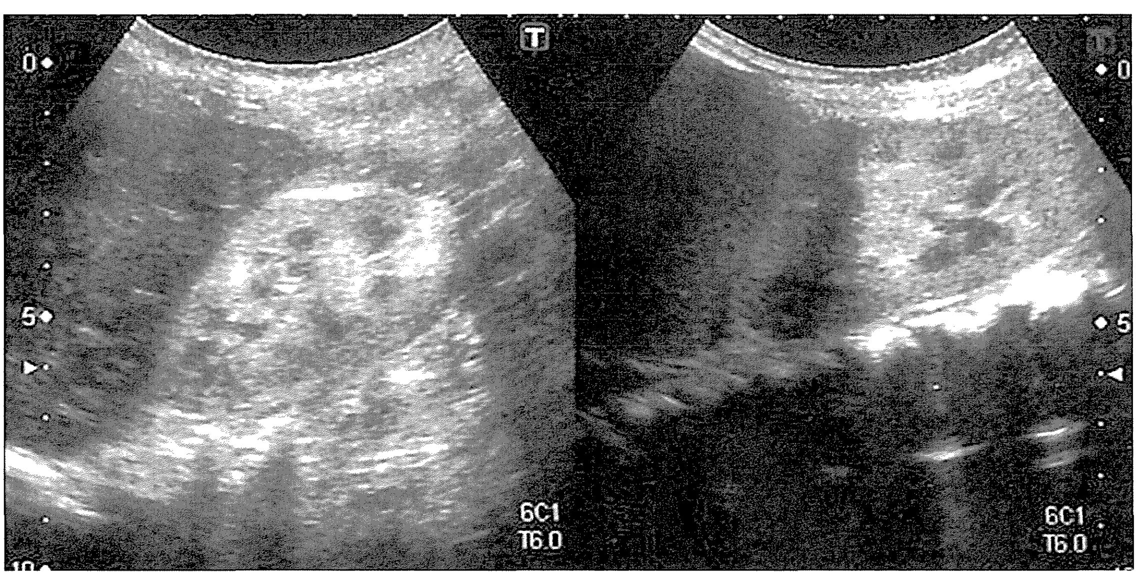

Fig. 5 Increased echogenicity of renal ultrasound in a patient with HUS

a European survey, 60 % of 394 patients with HUS received renal replacement therapy. Renal replacement therapy started usually on the 10th day in the USA [2] and on the 12th day in Japan [1] in average after STEC gastrointestinal infection manifested. HUS patients develop oliguria, anuria, edema, proteinuria, hematuria (including macrohematuria) and urinary casts. They also manifest an increase of serum creatinine, hyperkalemia, and hyponatremia. There are two main pathophysiology of AKI due to HUS, prerenal failure and intrinsic renal failure. The cause of intrinsic renal failure is STX which mainly affects the endotherial cells. STX is composed of an A subunit with toxic activity, and five B subunits with cell binding activity. The B subunits bind to Gb3 receptor (globotriosylceramide 3 receptor) of target cells. Upon binding, only A subunit is transported to cytoplasm. RNA N-glycosidase activity of A subunit inhibits 60S ribosome. Finally, the A subunit irreversibly inhibits protein production, leading to cell death. Gb3 receptors are expressed on red blood cells, white blood cells and endotherial cells. Renal interstitium is also a frequent target, as renal tubules also express Gb3 receptors [a].

Ultrasound findings of kidneys in HUS patients (in oliguric or anuric period disclose) include normal or enlarged kidney, hyperechoic renal cortex (higher than liver) and hypoechoic medulla (pyramides renales) [b]. Ultrasonography also revealed the absence of peripheral arterial diastolic blood flow or significant decrease of peripheral arterial diastolic blood flow in the kidneys. In the recovery period, peripheral arterial diastolic blood flow gradually increased.

Pathologic findings revealed narrowed lumen of glomerular capillary due to the endothelial edema and thrombus formation in the glomerular capillaries. Swollen and degenerated mesangial cells leading to mesangiolysis were also observed. Glomerulus was collapsed by thrombus of interlobular artery and arteriole, leading to fibrinoid necrosis. When thrombus formation in the interlobular artery woul be widespread, cortical necrosis could ensue. Inflammatory cytokines and chemokines such as TNF-α, IL-1β, IL-6 and IL-8, complement system and coagulation system including Von Wilbrand factor are involved in endotherial cell injury. Hypovolemia is an exacerbating factor of AKI in HUS [c, d].

2. Risk factors of AKI

Prerenal failure can contribute to cause AKI in patients with HUS. Renal replacement therapy was frequently needed in patients with HUS who manifested hypovolemia on admission [3, 4]. HUS patients who received insufficient fluid (water and sodium) therapy often manifest oliguria or anuria. Such patients frequently receive renal replacement therapy (refer to Sect. 3.1) [3, 4]. When patients with STEC infection or potential HUS patients first present themselves to the doctor, it is important to assess the dehydration level and to confirm how much of which kind of fluid is to be administrated in anticipation of AKI. In a nationwide survey conducted in Japan, indication of renal replacement therapy is 64 % (odds ratio 8:1) for patients who manifest hyponatremia (under 130 mEq/L) at onset of HUS, and 73 % (odds ratio 8:1) for patients who manifest elevated serum ALT over 70 IU/L [1]. Hospital induced hyponatremia is often introduced when hypotonic fluid was administered to STEC patients. Iatrogenic hyponatremia can cause brain edema leading to acute encephalopathy. Therefore, it is mandatory to evaluate serum electrolytes and water balance in patients with possible STEC infection. Specific serotype in STEC can cause severe form of HUS. Patients with O157:H7 infection are more likely to receive renal replacement therapy and manifest bloody stool (with elevated serum LDH) than patients with O157:non-H7 infection [2].

3. Staging and timing of renal replacement therapy in AKI

It is necessary to access serum creatinine and urine volume in patients with HUS who manifest AKI in order to determine the necessity of renal replacement therapy. Kidney Disease Improving Global Outcomes (KDIGO) disclosed clinical diagnostic criteria for AKI (Table 6) [e]. AKI stage is categorized by elevated value of serum creatinine and urine volume. Standard serum creatinine values for age and gender may vary as these values depend on the quantity of muscle (See Sect. 2.1).

Renal replacement therapy is indicated when signs of fluid overload (including pulmonary edema, cardiac failure, hypertension, electrolyte abnormality, hyperkalemia, hyponatremia and acidemia, nausea, vomiting, consciousness disorder and convulsion) are seen. The list of signs, however, is by no means exhaustive. Renal replacement therapy should be prepared before patients progress to AKI, which is potentially life-threatening. The KDIGO

Table 6 Staging of AKI [d]

Stage	Serum creatinine	Urine output
Stage 1	1.5–1.9 times baseline* or ≥0.3 mg/dL (≥26.5 μmol/L) increase	<0.5 mL/kg/h for 6–12 h
Stage 2	2–2.9 times baseline*	<0.5 mL/kg/h for 12 h
Stage 3	3 times baseline* or increase in serum creatinine to ≥4.0 mg/dL (≥353.6 μmol/L) or inhibition of renal replacement therapy or <18 years, decrease in eGFR to <35 mL/1.73 m^2	<0.3 mL/kg/h for ≥24 h or Anuria for ≥12 h

* Assess within 7 days

guidelines for AKI recommend that patients with Stage 2 AKI (and over two times of normal serum creatinine level, or urine volume less than 0.5 mL/kg/h for 12 h) should be transferred to medical institutions where renal replacement therapy and critical care medicine are available.

Supplementary articles

a. UpToDate: Clinical manifestations and diagnosis of Shiga toxin associated (typical) hemolytic uremic syndrome in children. (Accessed on December 1, 2012)
b. Siegel MJ: Urinary tract. In: Siegel MJ. (ed), Pediatric Sonography. Lippincott Williams & Wilkins, Philadelphia, pp.385–473, 2002.
c. Johnson S, et al. Hemolytic uremic syndrome. In: Avner E, Harmon WE, Niaudet P, Yoshikawa N. (eds), Pediatric Nephrology 6th ed. Springer-Verlag, Berlin Heidelberg, pp. 1155–1181, 2009.
d. Laszik ZG, et al. Hemolytic uremic syndrome, Thrombotic thrombocytopenic purpura and other Thrombotic microangiopathies. In: Jennette JC, Olson JL, Schwartz MM, Silva FG. (eds), Heptinstall's Pathology of the Kidney 6th ed, Wolters Kluwer, Lippincott Williams and Wilkins, Philadelphia, pp. 701–764, 2007.
e. Kidney Disease Improving Global Outcomes (KDIGO) Acute Kidney Injury Work Group. KDIGO Clinical Practice Guideline for Acute Kidney Injury. Kidney Int. Suppl 2012;2:1–138.

2.3 Diagnosis of encephalopathy

> It is not uncommon for STEC infection to be complicated by acute encephalopathy, immediately before or after the onset of HUS. Common clinical symptoms include convulsion and impairment of consciousness. On suspicion of encephalopathy (when probable diagnostic criteria are met), cranial imaging studies (CT or MRI) and electroencephalography (EEG) should be performed. [Grade of Recommendation: Not Graded]
>
> Diagnostic criteria
> Definite:
> Presence of one of the following findings during the course of STEC infection.
> (1) Clinical signs of convulsion and/or impairment of consciousness. Abnormal imaging findings (bilateral deep gray matter lesions or diffuse brain edema) on cranial CT or MRI.
> (2) Impairment of consciousness (equal to or above II-10 on Japan Coma Scale, or equal to or below 13 on Glasgow Coma Scale) lasting for more than 24 h.
>
> Probable:
> Clinical signs of convulsion and/or impairment of consciousness during the course of STEC infection.

Comments

1. Central nervous system (CNS) involvement and HUS in STEC infection

STEC infection is often complicated by CNS involvement as well as HUS. In the paper describing HUS for the first time in 1955 [1], CNS involvement was regarded as a part of HUS; whereas many papers after 1970 dealt with CNS involvement as an extrarenal complication independent of HUS. However, the vast majority of patients with CNS involvement also have severe HUS. Patients who developed CNS involvement and expired before meeting the diagnostic criteria of HUS have been reported, but are only exceptional [2]. Signs of CNS involvement often appear just after the onset of HUS (between 24 to 48 h). Approximately 10 % of HUS cases have CNS involvement, although the ratio ranges from 5 % to more than 30 % according to the studies [3–5].

2. Encephalopathy associated with STEC infection

In the acute stage of HUS, there are variable CNS signs, including convulsion (generalized or partial), impairment of consciousness (coma, stupor and hallucination), hemiparesis and decerebrate posture. Convulsion and impairment of consciousness are noted in more than 50 % of HUS patients [3, 5, 6]. An early but tentative diagnosis of "suspicion of encephalopathy" can start therapy based on the clear evidence of STEC infection and on neurologic findings of convulsion and/or impairment of consciousness. Subsequently, when impairment of consciousness is severe (e. g. equal or above II-10 on Japan Coma Scale or equal or below 13 on Glasgow Coma Scale) and long in duration (more than 24 h), a definite diagnosis of acute encephalopathy can be made.

CT or MRI, and EEG are useful for diagnosis. Although mild cases may appear normal on CT and MRI, severe cases often show diffuse brain edema and/or bilateral deep gray matter lesions (basal ganglia or thalami) (Fig. 6) [4, 7–9]. EEG reveals abnormal basic activity (an increase in slow wave) even in mild cases, and a more pronounced increase in slow wave and abnormal paroxysmal activity in severe cases [10].

Main pathogenesis is explained on the basis of systemic STX and inflammatory cytokines, causing dysfunction of cerebral blood vessels (in particular, increased permeability or breakdown of blood–brain barrier), together with the direct toxicity of intracerebral STX. Abnormalities in water, electrolytes and circulation due to acute renal injury (indicating hypertension), may also be present in varying significance among cases [3, 11, 12].

3. Cerebral infarction associated with STEC infection

Some HUS patients develop cerebral infarction. The timing of its onset varies greatly from the acute phase to the convalescence of HUS. There are focal neurological signs,

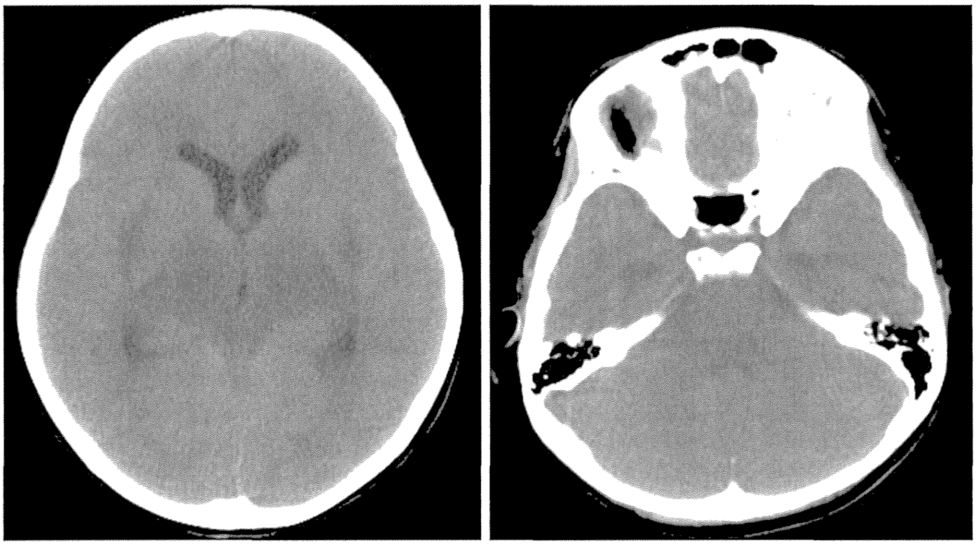

Fig. 6 Acute encephalopathy in an 8-year-old girl with STEC O157 infection (Cranial CT). Diffuse brain edema, together with low density and swelling of the bilateral thalami, putamina, external capsules and pontine tegmentum, can be seen

such as hemiparesis, ataxia and involuntary movements. Diagnosis is made on the basis of cranial CT and MRI, which visualize lesions varying from tiny lacunar infarcts to large hemorrhagic infarcts [13, 14]. Pathogenesis is explained mainly on the basis of thrombotic microangiopathy, with accompanying hemorrhagic diathesis due to thrombocytopenia and other factors described in the previous section.

2.4 Acute-phase extrarenal complication (excluding encephalopathy)

1. Hypertension

> Hypertension may occur during the acute phase of HUS. [Grade of Recommendation: Not Graded]

Comments

Hypertension occurs in up to 25 % of patients with HUS during its acute phase, resulting from overflow, renal glomerular disorder and vascular disorder, etc. [1–3] (See Sect. 3.2).

2. Gastrointestinal complications

> HUS patients infected with STEC will experience gastrointestinal complications such as marked gastrointestinal edemas, intussusception, rectal prolapse, appendicitis, intestinal necrosis and perforation, peritonitis, acute pancreatitis, and bile stasis or cholelithiasis. [Grade of Recommendation: Not Graded]

Comments

Marked intestinal edema is observed in HUS patients infected with STEC. Characteristic findings in abdominal ultrasonography are marked thickening of the ascending colon and enhancement of echo brightness [a]. Thickening extends from the ileocecal region to the anus, extending over the entire large intestine in severe cases [a]. Blood flow in the large intestinal wall decreases in the early phase of the disease and increases in the recovery period [a]. Reduced blood flow in the large intestinal wall observed in the early stage of the disease is caused by ischemia arising from microvessel fibrin thrombus [4].

In the mass infection among school children in Sakai City, Japan in 1996, several patients underwent appendicitis surgery for intestinal complication associated with STEC infection and those with intussusception were also reported [b]. Rectal prolapse was observed in 8 % of the children, while 3 % had concurrent intussusception [5]. However, marked large intestinal edemas sometimes occur in patients with gastrointestinal STEC infection, showing a target sign-like image in abdominal ultrasonography. Differential diagnosis from intussusception is required. Thickening of the large intestinal wall by STEC infection observed without pseudokidney sign may be used as a reference.

As patients with STEC gastrointestinal infection usually present with severe acute abdominal pain, they are sometimes misdiagnosed with acute appendicitis. However, when ultrasonography shows thicked wall of ascending colonin addition to the appendix, the diagnosis of appendicitis should be made under careful consideration [6].

Severe patients with HUS sometimes manifest necrosis, perforation, peritonitis of gastrointestinal tract and acute pancreatitis [7]. For diagnosis of acute pancreatitis, in addition to serum amylase level, its fraction and serum lipase should also be used as a reference. When renal function decreases, the level of urinary amylase secretion decreases while the level of serum amylase increases.

As a large volume of hemolysis occurs over a short period for patients with HUS, a transient retention of biliary sludge occurs in the gall bladder, which may lead to concurrent gallbladder stone [8–11]. Furthermore, it can cause biliary tract infection, acute pancreatitis, and liver function impairment [5, 7].

Supplementary articles

a. Sivit CJ, et al. Gastrointestinal tract. In: Siegel MJ. (ed), Pediatric Sonography. Lippincott Williams & Wilkins, Philadelphia, pp. 337–385, 2002
b. Task Force on the Mass Outbreak of Diarrhea in Schoolchildren of Sakai City: 1997. Sakai City, Japan: Sakai City Medical Association; Sakai City, Japan: Sakai City Medical Association.

3. Diabetes

Concurrent diabetes may occur in the acute phase of HUS as a result of decreased insulin secretion.[Grade of Recommendation: Not Graded]

Comments

In the acute phase of HUS, thrombosis of nutrient vessels of the pancreas may cause necrosis, inflammation, fibrosis of pancreatic artery or islet, and reduced insulin secretion, leading to the onset of diabetes. Its frequency is 1.7 % [12] to 3.2 % [a]. Patients requiring dialysis or patients with central neurological symptoms are prone to the onset of diabetes.

Supplementary article

a. Sivit CJ, et al. Gastrointestinal tract. In: Siegel MJ (ed), Pediatric Sonography. Lippincott Williams & Wilkins, Philadelphia, pp. 337–385, 2002.

4. Cardiovascular complications

During the acute phase of HUS, myocarditis, cardiac microthrombosis, dilated cardiomyopathy, cardiac tamponade, myocardial ischemia can occur. Please note that this list of conditions is not limited to the ones stated here. [Grade of Recommendation: Not Graded]

Comments

Myocarditis, cardiac microthrombosis, dilated cardiomyopathy, cardiac tamponade, myocardial ischemia, etc., can occur during the acute phase of HUS [a, 13–15]. In one patient with HUS who passed away suddenly during the acute phase [16], pathological findings disclosed inflammatory cellular infiltration in the cardiac muscle and the surrounding area of the conducting path.

Supplementary article

a. Siegler R. Cardiovascular involvement in the hemolytic uremic syndrome. In: Kaplan BS, Trompeter RS, Moake JL. (eds), Hemolytic uremic syndrome and thrombotic thrombocytopenic purpura. Dekker, New York, pp. 143–149, 1992.

3 Treatment of HUS

3.1 Fluid therapy and blood transfusion

1. Fluid therapy

We suggest sufficient intravenous fluid therapy using isotonic solutions be used for patients with STEC infection before the development of HUS. This is effective for preventing oliguria, anuria and dialysis. [Grade of Recommendation: C1] When a patient manifests oliguria or anuria, excessive fluid therapy may induce hypertension, lung edema or electrolyte abnormalities. For such patients, the volume of daily intravenous infusion should not exceed the total daily urinary output, insensible water loss and water loss from stool. [Grade of Recommendation: B]

Comments

Most patients with HUS develop AKI. Dehydration due to vomiting and diarrhea exacerbates AKI. Therefore, fluid therapy for dehydration is critical. On the other hand, patients with oliguria or anuria due to AKI are likely to develop hypertension, lung edema, electrolyte abnormalities and cardiac failure by excessive fluid therapy. For these patients, the volume of daily intravenous infusion should not exceed the total daily urinary output, insensible water loss and water loss from stool. During the acute phase of HUS, ingoing and outgoing balance of water, serum electrolytes and blood sugar should be frequently monitored.

Recent literature regarding patients with STEC infection has demonstrated that aggressive fluid therapy before the onset of HUS or in its early stages can prevent oliguria or anuria [1, 2]. A prospective cohort including 50 children with HUS showed that aggressive intravenous fluid therapy significantly reduced oliguria or anuria (fluid therapy group vs. non-fluid therapy group: 13/52 (52 %) vs. 21/25 (84 %), $p = 0.02$, odds ratio 1.6). Meanwhile, the non-oliguric/anuric group ($n = 16$) had received more total intravenous fluid and more total sodium than the oliguric/anuric group ($n = 34$) during the four days from onset of diarrhea; total volume of fluid and sodium was 1.7 (0–7.5) L/m^2 and 189 (0–483) mEq/m^2 in non-oliguric/anuric group and 0 (0–4.9) L/m^2 and 0 (0–755) mEq/m^2 in oliguric/anuric group, respectively (p value was 0.02 and 0.05).

Multivariable analysis demonstrated that the most significant covariate was total volume infused during the 4 days from onset of diarrhea. However, total volume and total sodium supplementation were also strong covariates [2].

A retrospective analysis of 137 children with HUS revealed that dehydrated patients on admission had higher risk of needing dialysis (70.6 vs. 40.7 %, $p = 0.0007$) [3]. In conclusion, aggressive fluid therapy using intravenous isotonic solution in the early stage of STEC infection or HUS may prevent AKI.

Anuria and dialysis in the acute phase of HUS are known to be risk factors of chronic renal damage such as albuminuria, proteinuria, hypertension and impaired renal function. Therefore, prevention of oliguria and anuria in the acute phase is significant from the viewpoint of long-term renal prognosis. Conversely, any potential protective effects against CNS involvement remained to be elucidated. Thus, for early intervention of intravenous fluid therapy, early detection of STEC infection is mandatory. Although various pathogens could cause gastroenteritis, the possibility of STEC infection should always be considered during examination of patients with gastroenteritis. If a patient has suspected STEC infection, stool culture or rapid diagnostic tests should be performed promptly.

Aggressive fluid therapy with isotonic solution in oliguric or anuric patients due to AKI may increase risks of hypertension, lung edema, electrolyte abnormalities and cardiac failure. Close monitoring of blood pressure, respiratory status and urination is necessary in such patients. Intravascular volume should be comprehensively evaluated by vital signs, cardiothoracic ratio on chest X-ray, aorta to inferior vena cava (IVC) ratio on ultrasound, and central venous pressure. Additionally, patients with HUS are likely to develop lung edema even with mildly increased intravascular volume as vascular endothelial injury increases vascular permeability. During the oliguric or anuric phase, supplementation of lost water and electrolytes (such as sodium and potassium) is a basic principle. In the clinical setting, many patients with STEC infection who have already progressed to HUS and AKI, developed iatrogenic hyponatremia resulting from continuation of hypotonic solution.

Some patients with HUS develop hyperkalemia due to AKI, while others develop hypokalemia as a result of severe diarrhea. In cases of severe hypokalemia, supplementation of potassium is required.

2. Blood transfusion

> We suggest that red blood cell transfusion be performed if hemoglobin dropped to <6 g/dL. [Grade of Recommendation: C1]
>
> To reduce the frequency of red blood cell transfusion, erythropoietin therapy from early phase of HUS can be considered. [Grade of Recommendation: C1]
>
> Platelet transfusion is usually not recommended as it may exacerbate thrombogenesis. However, in cases of severe bleeding tendency or massive bleeding, platelet transfusion is required. [Grade of Recommendation: C2]

Comments

Hematological complications of HUS include hemolytic anemia and thrombocytopenia. Primarily, only red blood cell (RBC) transfusion is allowed. However, this procedure should be used minimally because it accelerates generation of bilirubin and gallstones. Platelet transfusion is limited to cases of severe bleeding tendency, massive bleeding and invasive medical procedure, as it may accelerate thrombogenesis and exacerbate the clinical condition.

(1) RBC transfusion

We suggest that RBC transfusion to be considered when the hemoglobin level is <6 g/dL. In the acute phase of HUS, transfused RBC could promptly result in hemolysis. Therefore, normalization of hemoglobin is unnecessary. Excessive RBC transfusion should be avoided as it may induce cardiac failure, lung edema and gallstones [a]. RBC transfusion should be performed slowly and carefully since blood transfusion may increase intravascular volume rapidly and may induce hyperkalemia. In cases of hyperkalemia, washed RBC transfusion or use of a potassium removal filter may be necessary.

Target corrected value of hemoglobin is 8–10 g/dL. A small, randomized controlled trial revealed that erythropoietin therapy initiated from the early phase of HUS reduced the frequency of RBC transfusion [4]. Evaluation with a larger sized trial is expected in the future.

(2) Platelet transfusion

A retrospective cohort survey revealed the possibility that central venous (CV) and peritoneal catheters can be safely inserted without platelet transfusion. There was no significant difference in bleeding-related complications at insertion of a peritoneal dialysis catheter between patients who received platelet transfusion ($n = 22$; $3.76 \pm 2.19 \times 10^4/\mu L$, $p = 0.005$) and those who did not ($n = 51$; platelet before transfusion $6.48 \pm 3.88 \times 10^4/\mu L$). Simultaneously, partial omentectomy and insertion of a CV catheter were performed in both platelet-transfused group (omentectomy 45.5 %, CV catheter 90.0 %) and non-transfused group (omentectomy 43.1 %, CV catheter 91.50 %). This result suggests that some minor surgical procedures such as insertion of peritoneal dialysis or CV catheters, and omentectomy can be performed to eliminate the need for platelet transfusion [5].

Supplementary article

a. Up To Date: Treatment and prognosis of Shiga toxin associated (typical) hemolytic uremic syndrome in children. (Accessed on December 1, 2012)

3.2 Antihypertensive therapy

> Hypertension commonly occurs in HUS during acute phase. An amount of circulating blood (intravascular volume) should be evaluated correctly, and rationalization of blood pressure can be promptly attained with proper infusion, diuretic drug or antihypertensive agent, etc.
> [Grade of Recommendation: C1]
>
> Calcium channel blockers can be employed as first line therapy against acute hypertension.
> [Grade of Recommendation: C1]

Comments

Hypertension is common in HUS during the acute phase and can cause acute heart failure and posterior reversible encephalopathy syndrome. As such, prompt antihypertensive therapy is required. In evaluating blood pressure, normal values of blood pressure are set up for every gender and age group in children (Tables 7 and 8) [a].

The causes of hypertension in HUS are excess of intravascular volume and/or activation of RAS (renin angiotensin system) associated with renal ischemia [b].

Correct evaluation of intravascular volume is indispensable for appropriate supportive therapy. Intravascular volume is evaluated according to vital signs, cardiothoracic ratio (by chest roentgenography), the aorta and inferior vena cava ratio (by abdominal ultrasonography), central venous pressure, etc. In many cases, intravascular volume decreases (intravascular dehydration) when the patient's predominant symptoms are gastrointestinal before HUS develops. Initial fluid therapy by isotonic infusion may prevent onset of AKI. However, when renal failure progresses, intravascular volume may increase due to reduced urine output after HUS develops. In such instances, the infusion volume is required to be adjusted.

In order to maintain intravascular volume appropriately, appropriate fluid therapy and diuretic drugs are required against excessive intravascular volume. The first line agent for diuretics is furosemide: 1–5 mg/kg/dose intravenously. If the effect is insufficient, dialysis is needed. The first line agent of antihypertensives is an oral administration of a calcium channel blocker (nifedipine or amlodipine). Nicardipine infusion is appropriate for urgent cases when oral antihypertensive is not possible or ineffective (Table 9). Since inhibitors of RAS may cause decrease in renal blood flow, they do not qualify as a first line agent. However, when antihypertensive is insufficient, inhibitors of RAS may be used together with a calcium channel blocker. Inhibitors of RAS may be used for the renoprotection over a long-term period when hypertension or proteinuria persists after HUS subsided [1, b].

Supplementary articles

a. JCS Joint Working Group 2012. [Guidelines for drug therapy in pediatric patients with cardiovascular diseases]. Circulation Journal. 2012;76:167–187, in Japanese.
b. UpToDate: Treatment and prognosis of Shiga toxin associated (typical) hemolytic uremic syndrome in children. (Accessed on January 23, 2013)

3.3 Renal replacement therapy

1. Timing to initiate renal replacement therapy in AKI arising from HUS

> Indications to initiate renal replacement therapy are oliguria in tractable to therapy (urination: 0.5 mL/kg/hr for over 12 h), uremic manifestations, hyperkalemia ($\geq$6.5 mEq/L), hyponatremia (<120 mEq/L), acidemia (pH < 7.20), fluid overload, pulmonary edema, cardiac failure, hypertension and renal impairment (in which crystalloid solution, colloid solution, blood products, and drugs cannot used).
> [Grade of Recommendation: C1]

Comments

KDIGO Clinical Practice Guideline for AKI shows the standard value of oliguria in which renal replacement therapy is needed [a]. Other published indications for renal replacement therapy in AKI were also consulted [b–d]. Renal replacement therapy is also indicated when serum sodium is under 115 mEq/L. However, this value is too risky for patients with AKI due to HUS. Hence, serum sodium less than 120 mEq/L should be indicative of commencement of renal replacement therapy in HUS patients. Frequent, low-efficient, short hemodialysis (FLESHD) and arrangement of

Table 7 Definition of hypertension [a]

Normal blood pressure (BP)	Both systolic and diastolic BP are less than 90th percentile
Prehypertension	Systolic and/or diastolic blood pressure levels are greater than or equal to the 90th percentile but less than the 95th percentile, or if BP exceeds 120/80 mmHg (even if BP is less than the 90th percentile for gender, age, and height)
Hypertension	Systolic and/or diastolic blood pressure that is greater than or equal to the 95th percentile for gender, age, and height on three or more separate occasions. Stage 1: systolic and/or diastolic BP between the 95th percentile and 5 mmHg above the 99th percentile Stage 2: systolic and/or diastolic BP higher than 5 mmHg above the 99th percentile

Table 8 Blood pressure levels of 50th percentile height based on gender, age of children and adolescents in the USA [a]

Age	Boys			Girls		
(year)	90th	95th	99th	90th	95th	99th
1	99/52	103/56	110/64	100/54	104/58	111/65
2	102/57	106/61	113/69	101/59	105/63	112/70
3	105/61	109/65	116/73	103/63	107/67	114/74
4	107/65	111/69	118/77	104/66	108/70	115/77
5	108/68	112/72	120/80	106/68	110/72	117/79
6	110/70	114/74	121/82	108/70	111/74	119/81
7	111/72	115/76	122/84	109/71	113/75	120/82
8	112/73	116/78	123/86	111/72	115/76	122/83
9	114/75	118/79	125/87	113/73	117/77	124/84
10	115/75	119/80	127/88	115/74	119/78	126/86
11	117/76	121/80	129/88	117/75	121/79	128/87
12	120/76	123/81	131/89	119/76	123/80	130/88
13	122/77	126/81	133/89	121/77	124/81	132/89
14	125/78	128/82	136/90	122/78	126/82	133/90
15	127/79	131/83	138/91	123/79	127/83	134/91
16	130/80	134/84	141/92	124/80	128/84	135/91
17	132/82	136/87	143/94	125/80	129/84	136/91

Systolic/diastolic BP (mmHg)

sodium concentration in dialysate will be taken to correct the serum sodium in the patients.

Supplementary articles

a. KDIGO clinical practice guideline for acute kidney injury: Timing of renal replacement therapy in AKI. Kidney Int Suppl 2012;2:89–92.
b. Gibney N, et al. Timing of initiation and discontinuation of renal replacement therapy in AKI: unanswered key questions. Clin J Am Soc Nephrol 2008;3:876–880.
c. Bellomo R, et al. Indications and criteria for initiating renal replacement therapy in the intensive care unit. Kidney Int Suppl. 1998;66:S106–109.

Table 9 Antihypertensive agents for children

Generic name	Brand name	Dosage and administration
Nifedipine	Sepamit® fine granules 1 %	0.25–0.5 mg/kg/dose orally every 4–6 h. Maximum: 10 mg/dose or 3 mg/kg/day
Amlodipine besylate	Norvasc® Amlodin® Tablets/OD Tablets (2.5, 5 mg)	2.5 mg once daily in children ≥6 years. Dosage must be titrated according to age, body weight, and patient's response, but does not exceed the dosage for adult
Nicardipine hydrochloride	Nicardipine® *injection* (1 mg/ml)	Initial: 0.1–1.0 μg/kg/min; titrate dose according to blood pressure: rate of infusion may be increased every 15–30 min; maximum dose: 4–5 μg/kg/min
Enalapril maleate	Renivace® Tablets (2.5, 5, 10 mg)	Initial: 0.08 mg/kg/day every 24 h *in children ≥1 month*. Dosage must be titrated according to age, patient's response
Lisinopril	Longes® Tablets Zestril® Tablets (5, 10, 20 mg)	Initial: 0.07 mg/kg/dose once daily in children ≥6 years. Dosage must be titrated according to age, patient's response
Valsartan	Diovan® Tablets (20, 40, 80, 160 mg)	Initial: dose dependent upon patient weight: <35 kg: 20 mg; ≥35 kg: 40 mg/day every 24 h orally. Dosage may titrate to age, body weight, and patient's response up to a maximum dose of 40 mg < 35 kg

d. Palevsky PM: Clinical review: timing and dose of continuous renal replacement therapy in acute kidney injury. Crit Care 2007;11:232–237.

2. Modality of renal replacement therapy for patients with AKI arising from HUS

> Modalities of renal replacement therapy are peritoneal dialysis (PD), intermittent hemodialysis (HD) and continuous hemo-dia-filtration (CHDF), including continuous hemo-dialysis (CHD) and continuous hemo-filtration (CHF). [Grade of Recommendation: B]
>
> We suggest to take CHD For PD (for 24h) for patients with AKI complicated with acute encephalopathy. [Grade of Recommendation: C1]

Comments

We suggest to take PD, IHD or CRRT (such as CHD, CHF and CHDF) depending on the status of the patients and nature of the institution. Characteristics of PD, IHD and CRRT are shown in Table 10. However, there are no randomized control studies which modality shows most effective.

CHD and CHDF are usually the first line method for AKI due to HUS, because CHF shows lower efficacy than CHD or CHDF. CHD is usually selected when patient manifests AKI with no neurological involvement. On the other hand, when patient with HUS manifests acute encephalopathy, CHDF is chosen to reduce serum inflammatory cytokines. In cases of patients with unstable circulation, CHDF is preferred over CHD.

AKI guideline of KDIGO suggests CHDF or PD (for 24 h), rather than intermittent renal replacement therapy (IRRT such as IHD), for AKI patients with acute brain injury, increased intracranial pressure or generalized brain edema (Grade 2B: we suggest the Quality of Evidence Moderate) [a]. IHD can worsen neurological status by changing cerebral perfusion pressure. That is why CHDF or 24-h PD is taken. IHD can induce sudden disequilibrium syndrome or an increase of intracranial pressure (dialysis disequilibrium) [b]. Both types of therapy usually do not produce disequilibrium syndrome, brain edema or hypotension due to the slow removal of fluids and solutes [1]. The characteristics of modalities of renal replacement therapy are shown in Table 11.

Commercially available dialysates for patients with AKI contained low potassium (2 mEq/L), low magnesium (1 or 1.5 mEq/L) and no phosphorus. As CRRT is usually maintained for over 24 h, potassium, magnesium and phosphorus can be added to dialysates to maintain the appropriate serum levels in patients [c].

Supplementary articles

a. KDIGO clinical practice guideline for acute kidney injury: Modality of renal replacement therapy for patients with AKI. Kidney Int suppl 2012:107–110.
b. Davenport A: Continuous renal replacement therapies in patients with liver disease. Semin Dial 2009;22:169–172.
c. Sawada M, et al. Necessity of adding phosphorus and magnesium to dialysate in CHDF. J Jpn Soc Pediatr Dialysis Transplantation 2007;27:95–97 [in Japanese].

3.4 Plasma exchange therapy

Plasma exchange therapy has no beneficial effect to reduce nephropathy in HUS. [Grade of Recommendation: C2]

When plasma exchange therapy is performed, we must prevent fluid overload by using renal replacement therapy in the patients with HUS. [Grade of Recommendation: Not Graded]

Comments

For aHUS, plasma exchange therapy is the first line treatment. Conversely, plasma exchange shows no beneficial effect for patients with HUS due to STEC infection [1, 2, a] (See Sect. 3.6).

Plasma exchange therapy can increase plasma osmolality, as it promotes the migration of fluids from the third space to the intravascular space. Precise management of fluid, electrolytes and acid–base balance is mandatory. Rapid increase of intravascular volume leads to severe complications such as hypertension, cardiac failure, pulmonary edema and brain edema. When plasma exchange therapy is performed, fluid overload must be avoided by using renal replacement therapy in the patients with HUS [3]. We suggest plasma exchange therapy for infants with HUS in hospitals with adequate clinical experience.

Supplementary article

a. Michael M, Elliott EJ, Ridley GF, Hodson EM, Craig JC, Editorial Group: Cochrane Renal Group: Interventions for haemolytic uraemic syndrome and thrombotic thrombocytopenic purpura. 21 Jan 2009.

3.5 Antithrombotic therapy for HUS

We do not recommend administration of antithrombotics including heparin, dipyridamole, and urokinase for HUS patients, especially those without complications of disseminated intravascular coagulation (DIC). To date, there is no clinical evidence that it is beneficial. [Grade of Recommendation: D]

For patients with HUS complicated by DIC, it is reasonable to use agents including nafamostat mesilate, gabexate mesilate, recombinant human thrombomodulin alpha, or antithrombin (AT, formerly known as antithrombin III). [Grade of Recommendation: C1]

Comments

Differential diagnosis between HUS and DIC is usually difficult, and can only be made on the basis of results of examinations and careful observation of symptoms. DIC is a thrombotic microangiopathy resulting from activation of the coagulation system, accompanied by deposition of fibrin clots in the lumina of blood vessels, consumption of coagulation factors, and microangiopathic hemolysis. These in turn lead to thrombocytopenia, a decreased plasma fibrinogen level, prolongation of prothrombin time (PT) and activated partial thromboplastin time (aPTT). HUS, on the other hand, is a thrombotic microangiopathy resulting from primary platelet activation due to primary endothelial injury, with the levels of coagulation factors, PT and aPTT all within the normal range. Therefore, the differences in coagulation abnormalities between these two disorders usually permit them to be distinguished [a].

Four prospective trials have compared combination of supportive care with antithrombotic agents (e.g. either urokinase and heparin, or dipyridamole and heparin) or supportive care alone [1–4]. However, thrombocytopenia, microangiopathic hemolytic anemia and the duration of renal failure were similar in both the control and treated groups. Accordingly, these studies did not demonstrate any advantages of these treatments, nor the comparison between streptokinase therapy and heparin therapy with supportive care alone. Hemorrhagic complications were more common in the group treated with streptokinase. Therefore, antithrombotic therapy has not been deemed suitable for use in HUS patients.

On the other hand, HUS patients complicated by DIC are often reported [5, 6]. To date, the published data are insufficient to allow any conclusions to be drawn about the efficacy or safety of treatment for HUS complicated by DIC. Therefore, we suggest administration of agents such as nafamostat mesilate, gabexate mesilate, recombinant human thrombomodulin alpha and antithrombin, in accordance with the diagnostic and therapeutic guidelines for HUS caused by enterohemorrhagic *Escherichia coli* infection (established by the Japanese Society for Pediatric Nephrology) [b], as well as expert consensus based on evidence for the treatment of DIC due to infection (established by the Japanese Society for Thrombosis and Hemostasis) [c]. However, it is important to note that hyperkalemia may occur during treatment with nafamostat mesilate, and that any bleeding tendency can be exacerbated by treatment with antithrombin. Further, it is noteworthy that the established data are insufficient to allow any conclusions to be drawn about the efficacy of recombinant human thrombomodulin alpha for treatment of symptoms of HUS, including encephalopathy, although this treatment may be effective for DIC.

Supplementary articles

a. UpToDate: Treatment and prognosis of Shiga toxin associated (typical) hemolytic uremic syndrome in children. (Accessed on April 17, 2012)
b. The Japanese Society for Pediatric Nephrology [Diagnostic and therapeutic guideline for hemolytic uremic syndrome associated with enterohemorrhagic *Escherichia coli* infection (revised version)], in Japanese. http://www.jspn.jp/gakujyutsu.html
c. Maruyama I, et al. Scientific Standardization Committee of the Japanese Society on Thrombosis and Hemostasis: Expert consensus based on the evidence for the treatment of disseminated intravascular coagulation arising from intravascular infection. Jap J Thrombo Hemost. 2009;20:77–113, in Japanese.
d. Aoki N, et al. Revision of examination data and findings for diagnosis of disseminated intravascular coagulation. Research Report 1988: 37–41. Grant-in-Aid for Research of Special Disease of Blood Coagulation Abnormalities from the Ministry of Health and Welfare of Japan, in Japanese.

Table 10 Characteristics of PD, IHD, and CRRT

	PD	IHD	CRRT
Duration	Continuously for 24 h	Intermittent	Continuously for 24 h
Simplicity	Technically simple	Technically complex	Technically more complex
Influence on hemodynamics	Small	Large	Small
Control of the removed fluids	Not accurately	Accurately	Accurately and easily
Anticoagulation	Not necessary	Necessary	Necessary
Disequilibrium syndrome	No	Yes	No
Catheter related trouble	Obstruction, fluid leak, and peritonitis	Hemorrhage, thrombosis, and sepsis	Hemorrhage, thrombosis, and sepsis
Availability to infant	Available, good indication	Available	Available but depend on the institute activity
Restraint to the patients	Not necessary	Necessary	Necessary

PD Peritoneal dialysis, *IHD* intermittent hemodialysis, *CRRT* continuous renal replacement therapy

3.6 Treatment of encephalopathy associated with STEC infection

1. Supportive therapy for encephalopathy associated with STEC infection

> Supportive therapy includes the basic treatment of encephalopathy associated with STEC infection. To suppress brain edema and seizures (convulsions), management of systemic organs and treatment of the central nervous system (CNS) signs are critical. The former aims to stabilize circulation and respiration; while the latter, to treat seizures (convulsions) and to lower the intracranial pressure. [Grade of Recommendation: C1]

Comments

(1) Factors to be considered in the treatment of encephalopathy associated with STEC infection.

Main symptoms of encephalopathy associated with STEC infection are seizures (convulsions) and impaired consciousness. Many of the severe cases show diffuse brain edema and/or bilateral deep gray matter lesions (basal ganglia or thalamus) on cranial imaging studies (CT or MRI) (See

Table 11 Applications of renal replacement therapy in various complications

Complications	Modalities	Reasons of indication
Unstable circulation	CRRT · PD	Both can prevent hypotension
Hyperkalemia	IHD	IHD can reduce plasma potassium concentration rapidly
	CRRT	CRRT cannot rapidly normalize hyperlkalemia compared to IHD. Iit is recommended to infants
Acute encephalopathy/ increased intracranial pressure	CRRT · PD	Both are preferred to IHD. Both can keep the intracranial pressure stable
Bleeding tendency	PD	Anticoagulation is not necessary. PD will not accelerate bleeding tendency
	IHD	Treatment time of IHD is shorter than that of CRRT, resulting in less use of anticoagulants
Severe fluid overload	CRRT	CRRT can enable precise and continuous removal of fluid
During mechanical ventilation	CRRT	CRRT easily and accurately can control fluid balance. CRRT is also helpful to restore from mechanical ventilation
Intestinal perforation	CRRT	PD can not be used in patients with intestinal perforation

PD Peritoneal dialysis, *IHD* intermittent hemodialysis, *CRRT* continuous renal replacement therapy

Sect.2.3). To correct the pathology and pathogenesis for such cases, supportive therapy is performed. Since neither randomized nor case–control studies with regard to the treatment of encephalopathy associated with STEC has been done to date, there is no report of any therapy that has a high degree of evidence. It should be reasonable, however, to apply a therapeutic strategy similar to that against acute encephalopathy associated with influenza and other viral infections [a].

It should be kept in mind that, in cases of encephalopathy associated with STEC infection, most patients have acute kidney injury associated with HUS, prompting the consideration for water overload, electrolytes imbalance, as well as changes in blood concentration of drugs due to hemodialysis. Secondary injuries to other organs, such as the liver and heart, may also occur, although they are less serious compared to the some severe cases of influenza-associated encephalopathy. Taken together, encephalopathy associated with STEC infection differs in several aspects from other encephalopathies.

While encephalopathy associated with STEC is the main cause of death in HUS [1], there have been several case reports that described patients recovering after several weeks of coma [2, 3]. Thus, active and continuous therapy should be kept in mind.

(2) Supportive therapy of encephalopathy associated with STEC infection

In the acute period of encephalopathy, principle of treatment consists of several aspects. First, management of systemic organs should be done vigorously. Circulation and respiration should be continuously monitored and stabilized by hydration, drug therapy, dialysis and mechanical ventilation. Carbon bicarbonate concentration in arterial blood should be maintained at the normal range, and the volume of body fluid managed adequately to avoid both overhydration and dehydration. Abnormalities of the body fluid components, such as serum electrolytes and glucose, if any, should be corrected.

Second, CNS signs and symptoms should be treated. The level of consciousness and seizures (convulsions) should be monitored continuously. Seizures are to be halted primarily with intravenous antiepileptic drugs. Many patients respond to benzodiazepines (diazepam, midazolam and others), whereas some intractable cases have clusters of seizures or status epilepticus, necessitating intravenous injection of a large amount of barbiturates (thiopental and others). To prevent the recurrence of seizures, antiepileptic drugs (midazolam, phenobarbital, fosphenytoin and others) are given with monitoring of their blood concentration. Attention should be paid to seizures due to hyponatremia and other abnormalities of electrolytes, as well as hypoglycemia. Treatment of increased intracranial pressure includes sedation and hyperosmolar therapy (glycerol and fructose). Mannitol is not recommended for encephalopathy associated with HUS, since this is excreted via kidneys and may aggravate renal failure. Monitoring of intracranial pressure should be considered in severe cases. Cooling must be introduced to the patients with hyperthermia [a, 4].

(3) Follow-up during convalescence and after discharge

During convalescence, patients should undergo cranial imaging studies, electroencephalography and, if necessary, developmental tests to check for residual abnormalities, immediately before or after discharge. If a patient is left with disabilities in intellect, higher cortical and motor function, or epilepsy, treatment and rehabilitation should be started for each condition. Even if no apparent sequelae were noted at discharge, learning disabilities or behavioral problems may manifest later in life. Thus, a long-term follow-up on the mental development is required.

Supplementary article

a. Morishima T, et al. Guideline for influenza encephalopathy: Revised edition. Jpn J Pediatr. 2009;62:2483–2528. [in Japanese]

2. Specific therapy of encephalopathy associated with STEC infection

> Many cases of encephalopathy associated with STEC have an unfavorable prognosis. To date, no specific therapy has been established for this condition. [Grade of Recommendation: Not Graded]
>
> Methylprednisolone (mPSL) pulse therapy may be considered in patients with severe STEC-associated encephalopathy whereby poor outcome is predicted with regard to neurologic function and/or survival. However, its efficacy has not established. [Grade of Recommendation: Not Graded]
>
> Plasma exchange may be considered in patients with STEC-associated encephalopathy (when its safety is verified) although its efficacy has not been established. This therapy should be performed in a medical facility with adequate experience. [Grade of Recommendation: Not Graded]

Comments

Encephalopathy (CNS involvement) associated with STEC infection is recognized globally to be a predictor of poor outcome. The management of encephalopathy is primarily based on systemic supportive care [a]. Although no specific interventions have been shown to be efficacious, there may be a role for mPSL pulse therapy, plasma exchange and other treatments. The efficacy of these agents is still unclear due to the small number of clinical experience reports.

(1) Methylprednisolone (mPSL) pulse therapy

To date, there are no studies that have evaluated the efficacy of mPSL pulse therapy for STEC-associated encephalopathy. There are only a few case reports in Japan. To determine whether steroids could be of clinical benefit in the treatment of HUS, Perez et al. [5] conducted trials of mPSL (5 mg/kg/day over seven days). They reported that there was no significant difference between the treatment groups in terms of the number of convulsive episodes or transfusion requirements.

In April 2011, an outbreak of STEC O111 infection occurred principally in Toyama prefecture, Japan. During this outbreak, mPSL pulse therapy was introduced for encephalopathy. 20 children with STEC O111 infection were eventually identified with eight of them developed encephalopathy. Three children without mPSL pulse therapy died, while all five children treated with mPSL therapy survived without neurological sequelae [b]. However, there is still insufficient evidence to establish the effectiveness of mPSL pulse therapy for STEC-associated encephalopathy (as other interventions, such as plasma exchange, were used concurrently).

STEC–HUS is systemic disorder characterized by thrombotic microangiopathy (TMA). However, postmortem examination of brain tissue has shown little evidence of TMA. Affected patients often have generalized cerebral edema and enlarged spaces around blood vessels that indicate increasing permeability [b]. Previous studies showed that the pathogenesis of STEC-associated encephalopathy involves inflammatory cytokines such as TNF-α and IL-6 [6, 7]. Moreover, two cases of HUS complicated by acute necrotizing encephalopathy (ANE) were reported [8]. The efficacy of mPSL pulse therapy has been established for ANE. Although there were no obvious side effects of mPSL pulse therapy in the STEC O111 outbreak, this therapy should only be administered for STEC-associated encephalopathy when close attention is paid to the potential side effects, such as infections, thrombus formation and hypertension.

In spite of the treatment of STEC-associated encephalopathy with mPSL pulse therapy not being established, we suggest that it can be considered in patients with severe STEC-associated encephalopathy (with the safety of the patients ensured as a prerequisite).

(2) Plasma exchange therapy

Plasma exchange therapy is sometimes used to treat severe STEC–HUS (especially when there is CNS involvement) based upon the reported benefits of plasma exchange in adults with TTP. Dundas et al. [9] reported that five of 16 adult cases (31 %) treated with plasma exchange died, while five of the six cases without plasma exchange (83 %) died. Nathanson et al. recently investigated 52 patients with severe initial neurological involvement associated with D + HUS (HUS associated with diarrhea). Eleven patients were treated with plasma exchange within 24 h after the first presentation of neurological signs. However, the outcome of this group was not significantly different from that of the others who were not treated with plasma exchange [10]. Colic et al. reported that an earlier start of plasma exchange for five patients with STEC O104:H4 associated HUS reduced the lactate dehydrogenase concentrations more effectively than later treatment, possibly indicating that early therapy ameliorates the course of severe HUS [11].

The efficacy and mechanism(s) of plasma exchange for severe HUS and CNS involvement are currently unknown. Moreover, this therapy is associated with problems such as pulmonary edema, infection and the high cost of treatment.

While the treatment of STEC-associated encephalopathy with plasma exchange is not established, we suggest that it can be considered in patients with severe STEC-associated encephalopathy (with the safety of the patients ensured as a prerequisite).

(3) Other treatments

In a German outbreak of STEC O104 in 2011, it was reported that eculizmab, a monoclonal antibody against complement factor C5, was beneficial in patients with

STEC–HUS and CNS involvement [12]. In contrast, eculizmab did not show any efficacy in a cohort study by Menne et al. [13]. Recombinant human soluble thrombomodulin (rTM) has effects on the complement control and has anti-inflammatory properties. There was one report of a small number of patients that evaluated the efficacy of rTM in children with STEC–HUS [14]. More evidence is needed to establish a new therapeutic strategy for STEC-associated encephalopathy.

Supplementary articles

a. UpToDate: Treatment and prognosis of Shiga toxin associated (typical) hemolytic uremic syndrome in children. (Accessed on December 16, 2012).
b. Sata T (ed.): 2011 Annual Report of the Research Committee on Epidemiologic, Bacteriologic and Clinical Studies of Cases of Food Poisoning due to STEC/O111.

3.7 Renal sequelae of HUS

Renal sequelae of HUS are albuminuria, proteinuria, decreased renal function and hypertension.
About 20–40% of HUS patients developed chronic kidney disease (CKD), a risk factor of end-stage kidney disease and cardiovascular complications. We recommend patient monitoring by examining albuminuria, proteinuria and measurement of blood pressure according to the severity in the acute phase:

(1) At least for fifteen years in patients who needed dialysis in acute phase, or anuria for more than 6 days.
(2) At least for fifteen years in patients under two years old at the onset, whose peak serum creatinine was higher than or equal to 1.5 mg/dL.
(3) Throughout the life in HUS patients who are positive for albuminuria, proteinuria, decreased renal function, or hypertension during follow-up.
(4) For 5 years in patients without any of the above mentioned conditions or renal sequelae.

[Grade of Recommendation: B]

Renal biopsy in the acute phase for HUS patients is not recommended, because the bleeding risk is high and pathological findings of the acute stage do not correlate to the renal prognosis.
[Grade of Recommendation: C2]

Comments

1. Renal sequelae

The mortality rate in the acute phase of HUS is 2–6 % in western countries [1] and 1.6 % in Japan [2]. Of the fatal cases, 88 % occurred in the acute phase [3]. The mortality rate has improved markedly by advances in acute renal replacement therapy and pediatric intensive care compared to the 1980s [a]. About 40 % of the HUS patients develop anuria [1] and approximately 40 % [b]–60 % [1, c] of the HUS patients required dialysis in the acute phase. A national survey between 2000 and 2001 in Japan revealed that oliguria or anuria was seen in 47 % and dialysis in 27 % of the patients [2]. Most patients who needed dialysis in the acute phase recovered their renal functions. About 20–40 % of the HUS patients, however, developed chronic kidney disease (CKD) for prolonged periods [4]. Since CKD is a risk factor of end-stage kidney disease and cardiovascular complications, continuous management is needed.

Mortality rate was 9 % and rate of end-stage kidney disease was 3 % based on a meta-analysis of 49 articles that covered 3,476 HUS patients between 1950 and 2001 [5]. Of the 2,372 survivors who were monitored for more than a year, 25 % were complicated by renal sequelae. The symptoms and frequency are as follows: decreased renal function 15.8 % (GFR; 60–80 mL/min/1.73 m^2: 8 %, 30–59 mL/min/1.73 m^2: 6 %, 5–29 mL/min/1.73 m^2: 1.8 %), proteinuria 15 % and hypertension 10 % (multiple answer) [5].

Albuminuria, a more sensitive indicator of renal damage than proteinuria, is useful for early detection of CKD in HUS patients. The frequency of albuminuria at three years [6] and mild renal dysfunction at 6 years [7] after the onset of HUS was high compared to normal control.

Hypertension is the most prominent renal sequela in HUS patients [8]. About 25 % manifested hypertension in the acute phase, while about 10 % did so in chronic periods [5]. Hypertension can also manifest without other complications, but hypertension usually develops in patients with proteinuria and renal dysfunction [9]. Monitoring the ambulatory blood pressure measurement (ABPM) for 24 h can reveal occult hypertension [10].

The frequency of renal sequelae is high in HUS patients and long-term renal prognosis is not always good. Therefore, follow-up of patients according to their condition is necessary.

2. Predictive factor of prognosis for renal function and follow-up

The risk factors of renal sequelae include oliguric or anuric period and dialysis period in acute phase [11]. When anuric period is over 7–10 days, renal sequelae such as proteinuria, renal dysfunction and hypertension increase

[12–15]. In addition, renal sequela correlates to the period of dialysis [5, 10]. The renal function can be decreased in patients who needed dialysis for more than 5 weeks [5].

11–16 % of the patients with HUS manifested renal dysfunction (<80 mL/min/1.73 m^2) during the follow-up period [5]. Furthermore, proteinuria and renal dysfunction manifested after 5 years in patients whose serum creatinine level was higher than 1.5 mg/mL in the acute phase [16].

As 951 children with gastrointestinal STEC O157 infection with no HUS did not manifest hypertension or microalbuminuria, long-term follow-up was not needed for these patients [17].

On the basis of the evidence above, we recommend follow-up of patients with HUS by examining albuminuria, proteinuria, and measurement of blood pressure according to the severity of the disease in the acute phase.

(1) At least for 15 years in patients who needed dialysis in acute phase, or anuria for more than 6 days.
(2) At least for 15 years in patients under 2 years old at the onset, whose peak serum creatinine was higher than or equal to 1.5 mg/dL.
(3) Throughout the life in HUS patients who are positive for albuminuria, proteinuria, decreased renal function, or hypertension during follow-up.
(4) For 5 years for patients without any of the above mentioned conditions or renal sequelae.

Renal pathological findings in the acute phase cannot predict long-term renal prognosis. However, examination of the kidney during the sub-acute phase revealed that patients with cortical necrosis and glomerular microangiopathy (covering more than 50 % of liver area) showed poor long-term renal prognosis (average 18 years) [13]. The indication for renal biopsy in the acute phase is to assist diagnosis [d], and surgical renal biopsy must be considered in the patients with high-risk bleedings. For patients with severe renal dysfunction and persistent proteinuria after the acute phase, renal pathological findings can produce important information for proper treatment. Renal biopsy in the acute phase for HUS patients is not usually recommended.

Supplementary articles

a. Johnson S, et al. Hemolytic uremic syndrome. In: Avner ED, Harmon WE, Niaudet P, Yoshikawa N (eds), Pediatric Nephrology 6th ed. pp. 1155–1180, Springer-Verlag, Berlin, 2009
b. Remuzzi G, et al. The hemolytic uremic syndrome. Kidney Int. 1995;48:2–19.
c. Bakkaloglu SA, et al. Diseases of the Kidney and Urinary Tract in Children. Taal MW, Chertow GM, Marsden PA, Skorecki K, Yu ASL, Brenner BM ed, Brenner and Rector's The Kidney 9th ed. pp. 2622–2679, Elsevier, Philadelphia, 2012.
d. UpToDate: Treatment and prognosis of Shiga toxin associated (typical) hemolytic uremic syndrome in children. (Accessed on January 23, 2013)

3.8 Extra-renal sequelae in patients with HUS

Patients with HUS can have extra-renal sequelae: sequelae of digestive system, diabetes mellitus, neurological complications, behavioral and cognitive sequelae or cardiovascular sequelae. It is important to conduct follow-up for HUS patients for at least five years after the acute illness. Long-term special care and treatment should be directed to patients with specific sequelae after the acute phase of HUS. [Grade of Recommendation: B]

Comments

1. Sequelae of digestive system

Cholelithiasis, persistent pancreatitis and colon stricture were reported in HUS patients as extra-renal sequelae [1]. Cholelithiasis is related to hemolysis or the use of parenteral nutrition in the acute illness. Pancreatic microthrombi can cause exocrine cell death resulting in persistent pancreatitis. Hemorrhagic colitis can cause severe inflammation of bowels leading to bowel stricture or obstruction. The transverse and ascending colon are most frequently affected. Bowel resection was indicated in patients with bowel stricture or obstruction who manifested persistent abdominal pain and severe constipation intractable to medication [2].

2. Diabetes mellitus

Pancreatic microthrombi can cause islet cell death resulting in diabetes mellitus. The incidence of diabetes mellitus during the acute phase of HUS was 1.7–3.2 % [3, 4]. HUS patients with severe disease (including the need for dialysis and CNS symptoms) were more likely to develop diabetes mellitus. Among those who developed diabetes mellitus, one-third had permanent diabetes mellitus. Relapse of diabetes mellitus can occur years after the acute illness. Patients with STEC infections who do not manifest HUS will not have diabetes mellitus.

3. Neurological sequelae

Convulsion and impaired consciousness in the acute phase of HUS are associated with severe renal damage [5]. In contrast, HUS patients with seizure or impaired consciousness in acute phase can recover without permanent neurological complications. Neurological outcomes including epilepsy, hemiplegia, cortical blindness and psychomotor disturbance were reported [6, 7].

4. Behavioral and cognitive sequelae

Patients who recovered from the acute phase of HUS can manifest mild behavioral and cognitive sequelae [8]. In contrast, patients who did not manifest neurological complications in the acute phase of HUS did not manifest learning disability, behavioral disturbance and attention deficit [9].

5. Cardiovascular sequelae

Myocarditis, cardiac thrombotic microangiopathy, dilated cardiomyopathy, cardiac tamponade and ischemic myocardial involvement were reported as cardiovascular sequelae in HUS patients after the acute phase. However, the long-term outcome of these cardiac complications is not known [a, 10–12].

Secondary evidence

a. Siegler R. Cardiovascular involvement in the hemolytic uremic syndrome. In: Kaplan BS, Trompeter RS, Moake JL (eds), Hemolytic uremic syndrome and thrombotic thrombocytopenic purpura. Dekker, New York, pp. 143–149, 1992.

4 Diagnosis and treatment of HUS in adults

4.1 Diagnosis of HUS in adults

1. The diagnosis of adult HUS

> There are a variety of etiologies in HUS in adults. Possibilities other than STEC-associated HUS should be explored particularly when it occurs in the absence of bloody diarrhea. [Grade of Recommendation: Not Graded]

Comments

The etiologies of adult HUS differ from those in children. Most HUS are caused by secondary diseases such as thrombotic thrombocytopenic purpura (TTP) associated with ADAMTS13 (a disintegrin-like and metalloproteinase with thrombospondin type 1 motifs 13) abnormality and various disorders that lead to aHUS [1–4, a–f] (Table 1). Oftentimes, it is difficult to distinguish between HUS and TTP at the onset, and plasmapheresis without delay being considered for most patients. Hence, at the stage of tentative diagnosis, the abbreviation of TTP/HUS (or HUS/TTP) is often used to describe the syndromes. Typical HUS caused by STX, which represents more than 90 % of HUS in children, is seen in only 5–10 % of the TTP/HUS cases in adults [a, b]. Table 12 shows the incidence of various TTP/HUS causes reported in the Japanese registry that covers mainly secondary causes [f].

STEC-associated HUS is usually considered in adult patients if they present with hemorrhagic diarrhea. Otherwise, other causes of secondary TTP/HUS should be explored. It is noteworthy that non-hemorrhagic diarrhea may be seen in about 30 % of non-STEC-associated HUS. On the contrary, hemorrhagic diarrhea can be seen when patients manifest ischemic colitis or peptic ulcers.

As shown in Table 12, the etiologies of TTP and atypical HUS varied and should be investigated according to the patient history and findings (see Sect. 5) [g]. DIC and malignant hypertension, and scleroderma kidney sometimes resemble HUS, but are usually diagnosed separately [a, d].

Prognosis of HUS in adults depends on its causes but is generally worse in the elderly patients. It was reported previously that the magnitude of renal damages could predict patient survival [4].

2. Treatment of HUS in adults

> We recommend treatment of underlying diseases and systemic supportive care for adult patients with HUS according to the guidelines for children. [Grade of Recommendation: B]
>
> We suggest initiating plasmapheresis without delay in adult patients with severe HUS, even if the etiology has not been determined. [Grade of Recommendation: C1]
>
> We suggest plasma infusion when plasmapheresis is not immediately available in adult patients with severe HUS. [Grade of Recommendation: C1]

Comments

The basis of treatment for adult patients with HUS is supportive care with careful systemic management similar to that for children. In addition, treatment of underlying diseases is of particular importance in adult TTP/HUS. Supportive management encompasses fluid infusion, transfusion of blood and its components, nutritional care and management of AKI including dialysis therapy [g]. The prognosis of TTP/HUS used to be very poor decades ago. However, it has since improved tremendously with the progress in supportive cares and the prevalent use of plasmapheresis [5–9, h]. Indication of plasmapheresis includes TTP with ADAMTS13 abnormality and most cases of complement-mediated aHUS (except for those caused by membrane cofactor protein/CD46 mutation) [9, g–i]. HUS secondary to certain drugs (ticlopidine, clopidogril, quinine) and HIV may also be indicated.

In contrast, HUS secondary to disseminated malignancy and most cases of hematological stem cell transplantation and STEC are not indicated. Plasmapheresis should be avoided for invasive pneumococcus-derived HUS usually seen in children, as anti-Thomsen-Friedenreich IgM antibody in serum may induce hemolysis that could exacerbate the pathogenesis of HUS (see Sect. 5). In patients with HUS secondary to autoimmune diseases, or for refractory or severe cases, immunosuppressive therapy may be combined with plasmapheresis. Unfortunately, it will take some time before the etiology of HUS is clarified. Prognosis in such cases is extremely poor if the initiation of plasmapheresis is delayed even for one or 2 days. If the diagnosis of TTP and aHUS is highly suspected, we strongly recommend that plasmapheresis be initiated without delay even with no known etiologies. We suggest that patient serum be taken for the purpose of future diagnostic use. Plasmapheresis should be terminated immediately when the etiology has been revealed in which plasmapheresis is not indicated or contraindicated.

Plasmapheresis is to be performed daily at the beginning and continued until the platelet count in the blood has normalized. Thereafter, it should be arranged according to the platelet count in the blood and serum LDH level. Alternatively, plasma infusion may be considered when prompt plasmapheresis is not available [7]. It has been reported that platelet transfusion might induce formation of microvascular thrombosis, but an analysis of the data in Oklahoma TTP-HUS registry revealed no such effect [10]. Therefore, when the risk for bleeding from thrombocytopenia is relatively high, platelet transfusion can be employed after careful consideration.

In autoimmune diseases such as connective tissue disease, treatment with glucocorticoids and immunosuppressive drugs may be considered. The efficacy of rituximab is not established for TTP, but may be considered in refractory or relapsing cases with anti-ADAMTS13 antibody [h]. Antiplatelet agents have not been shown to be effective for TTP and aHUS [h].

Supplementary articles

a. UpToDate: Causes, Diagnosis, and Treatment of thrombotic thrombocytopenic purpura-hemolytic uremic syndrome in adults. (Accessed on July 24, 2012)
b. Noris M, et al. Hemolytic-uremic syndrome. J Am Soc Nephrol. 2005;16:1035–1050.
c. Clark WF, et al. Attending rounds: microangiopathic hemolytic anemia with renal insufficiency. Clin J Am Soc Nephrol. 2012;7:343–347.
d. Kagami S, et al. Diagnostic criteria of atypical hemolytic uremic syndrome. Nihon Jinzo Gakkai Shi. 2013;55:91–93.
e. Fujimura Y, et al. Registry of 919 Patients with Thrombotic Microangiopathies across Japan: Database of Nara Medical University during 1998-2008. Intern Med 2010;49:7–15.
f. George JN. How I treat patients with thrombotic thrombocytopenic purpura: 2010. Blood 2010;116:4060–4069.
g. Taylor CM, et al. On behalf of a working party from the Renal Association, the British Committee for Standards in Haematology and the British Transplantation Society: Clinical Practice Guidelines for the management of atypical Haemolytic Uraemic Syndrome in the United Kingdom. Br J Haematol. 2012;148:37–47.
h. Scully M, et al. On behalf of British Committee for Standards in Haematology: Guidelines on the diagnosis and management of thrombotic thrombocytopenic purpura and other thrombotic microangiopathies. Br J Haematol. 2012;158:323–335.

4.2 Diagnosis and treatment of STEC-associated HUS in adults

1. Clinical features of STEC-associated HUS in adults

> STEC-associated HUS may occur in an outbreak or sporadically in adults, although the incidence is lower compared to in children. [Grade of Recommendation: Not Graded]
>
> Elderly people with STEC infection are likely to develop HUS and prognosis is usually poor for such cases. [Grade of Recommendation: Not Graded]

Comments

STEC infection is seen in 5–10 % of adult TTP/HUS [a]. Sporadic and community-based infection may occur, with outbreaks being reported in elderly nursing homes. Currently, it is still not known why sporadic infection is seen more frequently in females together with slightly higher incidence of outbreak [1, 2]. O157 is the most common specie that causes HUS in adult, and the same is observed in children. However, O104 in Germany and O111 in Japan accounted for the majority of adult cases [b]. Other species such as O111, O145, O26 and O121 have been reported before [a]. The Oklahoma TTP/HUS registry showed that in comparison with children, 21 adult cases (21–89 years with a median of 59 years) showed more severe manifestations in the CNS, anemia, thrombocytopenia and poor prognosis, although the degree of AKI was similar between adults and children [1].

In the outbreak of O104 in Germany in 2011, there were almost no differences in clinical features between the cases with and without HUS. The incidence of HUS was lower in adults (average 37 years) than in childhood cases, although hemorrhagic diarrhea was seen more frequently [2]. The reason has not been elucidated; the differential expression of Gb3 receptor for STX in the intestines, which was suggested in animal studies, has not been examined in human cases of HUS [c]. For adults with HUS, it was seen mainly in females.

Outbreak of STEC infection may occur among elderly people in facilities like nursing home, and ages older than 65 years are reported as a risk factor for development of HUS in patients infected with STEC [3]. Therefore, elderly patients with STEC infection should be managed more carefully from the onset. If they present with HUS, systemic treatment should be initiated without delay. Possible explanations as to why elderly people with infection show poor outcome include decreased antibody titers against STX [4] and reduced defense mechanisms against infection in the stomach. The latter is most likely to be caused by reduced gastric juice secretion, gastrectomy and the use of antacid agents [3].

2. Treatment of adult HUS caused by STEC infection

> We recommend treatment of underlying diseases and systemic supportive care for adult patients with STEC-associated HUS according to guidelines for childhood cases. [Grade of Recommendation: B]
>
> We suggest plasmapheresis or combined therapy of immunoadsorption with IgG infusion in adult patients with severe STEC-associated HUS presented with CNS involvement. [Grade of Recommendation: C1]
>
> For STEC-associated HUS in adults, no evidence is present for the use of antibiotics and eculizumab. [Grade of Recommendation: Not Graded]

Table 12 Etiology and prevalence of HUS in adults [e]

TTP (ADAMTS13 deficiency and anti- ADAMTS13 antibody) 30–40 %
STEC-HUS 4–10 %
Others (atypical HUS) 50–60 %
Hereditary (abnormality of complement-regulated gene and others) No data available
Idiopathic
Drugs
Antiplatelet drugs: ticlopidine, clopidogril
Anticancer drugs: mitomycin C, gemcitabine
Calcineurin inhibitors: cyclosporine, tacrolimus
Quinine
Pregnancy (HELLP syndrome, pregnancy-associated hypertension, etc.)
Infection (HIV, streptococcus pneumonia, influenza virus, etc.)
Autologous hematological stem cell transplantation
Connective tissue disease (SLE, anti-phospholipid antibody syndrome, systemic sclerosis, etc.)
Malignancy (malignant lymphoma, gastric cancer, etc.)
Others

Comments

Basic management for adult patients with STEC-associated HUS is similar to that for childhood cases. It includes systemic supportive care such as fluid infusion, transfusion of blood and its components, nutritional care and management of AKI. In severe cases, intensive care with respiratory and circulatory management is mandatory. In some reports, plasmapheresis was shown to be beneficial for the improvement of patient survival [5, 6]. In the outbreak of O104 in Denmark in 2011, plasmapheresis was reported to be effective for patients with neurological disturbances showing consciousness loss or convulsion [6]. In contrast, plasmapheresis did not show any efficacy in the 2011 German outbreak of O104 [7]. These are contradicting reports with regard to the efficacy of plasmapheresis. Randomized controlled trials are therefore necessary to determine the efficacy and indication for plasmapheresis. Overall, we suggest performing plasmapheresis for severe patients with poor prognosis who have no other suitable treatment options.

It was reported recently that a combined therapy of immunoadsorption and IgG infusion was effective for 12 HUS patients with severe neurological disturbances [8], and that efficacy was observed even in the patients who were refractory to treatment with plasmapheresis. Although the study has a limited patient number, such combined therapy may be considered for adult patients with refractory cases of severe HUS with neurological disturbances. While it remains unknown which treatment of immunoabsorption and IgG infusion is more important, there was a report that IgG infusion alone was not effective in childhood cases [d].

There are several reports showing the use of antibiotics may worsen the prognosis of HUS patients. On the other hand, it has recently been reported that azithromycin may shorten the duration of bacteremia in adult patients with O104-associated HUS [9], although it did not affect renal and patient survival [9]. In the 2011 German outbreak of 104, it was reported that involvement of the CNS was less as a result of antibiotics use. HUS patients treated with a multiple regimen of antibiotics has better prognosis than those who were not [7]. However, there are no other reports suggesting the usefulness of antibiotic therapy. As such, the efficacy of antibiotics remains to be clarified.

Eculizumab has been used for patients with STEC-associated HUS to suppress activated complement system, and showed good efficacy in childhood cases [10]. However, no efficacy was observed in the cohort study of adult cases (average age 47.7) in the O104 outbreak shown above [7]. We have decided not to show recommendation grade for antibiotics and eculizumab. More evidence is clearly required to determine its efficacy.

Supplementary articles

a. Noris M, et al. Hemolytic-uremic syndrome. J Am Soc Nephrol. 2005;16:1035–1050.
b. Sata T. Epidemiologic, microbiological and clinical research in cases of EHEC/O111 food poisoning. Research Report 2011, Grant-in-Aid for Scientific research from the Ministry of Health, Labour and Welfare of Japan.

c. Mobassaleh M, et al. Developmentally regulated Gb3 galactosyltransferase and a-galactosidase determine Shiga toxin receptors in intestine. Am J Physiol. 1994;267:G618–G624.
d. Remuzzi G, et al. The hemolytic uremic syndrome. Kidney Int. 1995;48:2–19.

5 Diagnosis and treatment of atypical hemolytic uremic syndrome (aHUS)

5.1 The diagnosis of aHUS

aHUS is a type of HUS characterized by a triad of microangiopathic hemolytic anemia, thrombocytopenia and AKI; and excludes STEC-associated HUS and TTP caused by markedly decreased ADAMTS13. [Grade of Recommendation: Not Graded]

Diagnostic criteria
Definite:
Definitive diagnosis of aHUS based on the presence of the complete triad, but an absence of STEC infection and TTP caused by a marked decrease of ADAMTS13.

1. Microangiopathic hemolytic anemia: the level of hemoglobin (Hb) is less than 10 g/dL (We defined microangiopathic hemolytic anemia as an Hb level of less than 10 g/dL. At diagnosis, the presence of microangiopathic hemolysis should be confirmed on the basis of reference data including elevation of LDH level, a markedly decreased serum haptoglobin level, and the presence of schistocytes in blood smears.)
2. Thrombocytopenia: a platelet count of less than 150,000/μL
3. AKI in pediatric case: a serum creatinine level exceeding 1.5-fold the reference value by age and gender issued by the Japanese Society for Pediatric Nephrology.

Probable:
Probable diagnosis is based on the presence of two components of the triad with the exclusion of STEC infection and TTP caused by a marked decrease of ADAMTS13.

Comments

aHUS has been traditionally regarded as a disease concept that excludes STX-associated HUS, the most common form of HUS in children. aHUS is a heterogeneous disorder responsible for only 10 % of cases in children. An increased number of cases of aHUS have been reported to develop from the pathogenesis of HUS. In the present guidelines, we followed and adopted the diagnostic criteria established by the Joint Committee of the Japanese Society of Nephrology and the Japanese Society of Pediatrics [a]. In view of the unreliability of diarrhea as a distinguishing feature, aHUS should be suspected if the following characteristics are present, irrespective of whether diarrhea is present: (1) Patient is less than 6 months of age, (2) disease recurrence, (3) latent onset, (4) familial history of the disease with food poisoning excluded. Classification of aHUS is shown in Table 13 [b]. For differential diagnosis of aHUS, examinations should be planned with understanding of the characteristics of the causative disease for HUS.

1. Invasive pneumococcal infection

Invasive pneumococcal infection is defined as severe pneumococcal disease manifested as severe pneumonia, meningitis, bacteremia, sepsis, empyema, and other conditions. The pathogenesis of pneumococci-associated HUS has been suggested to involve the release of *N*-acetylneuraminidase, which cleaves *N*-acetylneuraminic acid in the glycocalyx, resulting in the exposure of the Thomsen-Friedenreich antigen on red blood cells, platelets, and glomeruli. Thomsen-Friedenreich antigen is recognized by a natural IgM antibody normally present in plasma leading to polyagglutination of the patient's red cells and hemolysis [c]. For diagnosis of pneumococci-associated HUS, identification of *Streptococcus pneumoniae* is necessary by culture, as well as detection of Thomsen-Friedenreich antigen on red cells [d].

2. Disorder of regulatory components of the complement system

Dysregulatory changes in complement system components should be estimated through measurement of homolytic complement activity (CH50), assay of complement protein and complement regulatory protein, detection of auto-antibody against complement factor H (CFH), and measurement of membrane cofactor protein (MCP, CD46) expression level on monocytes [e]. Thereafter, genetic complement-associated HUS can be definitively diagnosed through gene analysis of complement proteins and complement regulatory proteins. However, missense mutations of complement proteins typically result in functional impairment without affecting serum complement protein levels [f]. Therefore, analysis of known candidate genes is recommended, if possible.

3. Deficiency of ADAMTS13

ADAMTS13 deficiency consists of two types, congenital type (Upshaw-Schulman syndrome) and acquired type due to its inhibitor, anti-ADAMTS13 antibody. A marked decrease of ADAMTS13 activity to a level of less than 5 % has been demonstrated in 60–90 % of patients with TTP. Therefore, patients with congenital or acquired TTP should be diagnosed and ruled by measuring the activity of ADAMTS13 and its inhibitor.

4. Cobalamine metabolism abnormality

Inborn error of cobalamine C metabolism is a rare cause of HUS, especially in young infants (less than 6 months of age). The diagnosis is suggested by a marked increase of homocysteine and a decrease of methionine demonstrated by plasma amino acid chromatography.

5. Recessive mutation in diacylglycerol kinase ε (DGKE) gene

Mutations in diacylglycerol kinase ε (DGKE) gene were identified using exome sequencing in four patients with aHUS [g]. Most patients with DGKE gene mutation presented with aHUS in the first year of life show episodes of relapse before 5 years of age. It was reported that 13 (27 %) of 49 patients with aHUS in the first year of life had DGKE gene mutations and that three of six familial disease kindreds had these mutations. Affected individuals present with aHUS in the first year of life have persistent hypertension, hematuria and proteinuria (sometimes in the nephrotic range), and commonly show progression to CKD stage 4 and 5 by the second decade of life. Therefore, DGKE gene mutations should be suspected if characteristic symptoms such as hypertension, hematuria and proteinuria occur after recovery from aHUS attacks, and that there are no pathogenic mutations in known aHUS-related genes or antibodies against CFH.

6. HIV infection

Definitive diagnosis is performed by serological test for anti-HIV antibody.

7. Others

Definitive diagnosis is performed with various examinations including serological examinations for anti-nuclear antibody and anti-phospholipid antibody.

5.2 Treatment of aHUS

Treatment of aHUS includes supportive therapy for control of overall body conditions and specific therapy for the causative disease. [Grade of Recommendation: B]

(1) Pneumococcal-associated aHUS

Plasma therapy, including plasma exchange and plasma infusion with fresh frozen plasma, should be avoided in therapy for pneumococcal-associated HUS as plasma (which contains natural IgM-class antibodies against Thomsen-Friedenreich antigen) may aggravate hemolysis. It is preferable to transfuse washed RBC or platelets. [Grade of Recommendation: D]

(2) aHUS associated with complement dysregulation and other abnormalities

The guideline indicates that plasma therapy, including plasma exchange and plasma infusion, should be started as soon as possible at diagnosis of aHUS (excluding cobalamine metabolism disorder and pneumococcal-associated HUS). [Grade of Recommendation: C1]

Patients diagnosed with aHUS (based on the diagnostic criteria proposed by the Joint Committee of the Japanese Society of Nephrology and the Japanese Society of Pediatrics) should be treated with eculizumab. [Grade of Recommendation: C1]

Living-related donor transplantation should not be performed in patients with end-stage renal disease (ESRD) due to aHUS. [Grade of Recommendation: C2]

Preventive plasma therapy should be performed in the perioperative period for patients with ESRD due to aHUS undergoing cadaveric unrelated renal transplantation. [Grade of Recommendation: C1]

Prophylactic eclizumab administration in the perioperative period is acceptable for patients with ESRD due to aHUS and undergoing cadaveric unrelated renal transplantation. [Grade of Recommendation: C1]

Table 13 Classification of aHUS (excluding TTP due to ADAM-TS13 deficiency)

1. Advanced Etiology
   - (i) Infection induced
     - *Streptococcus pneumoniae* infection
   - (ii) Disorders of complement regulation
     - Genetic disorders of complement regulation: complement factor H (CFH), complement factor I (CFI), membrane cofactor protein (MCP, CD46), C3, complement factor B (CFB), thrombomodulin
     - Acquired disorders of complement regulation: auto-antibody
   - (iii) Defective cobalamine metabolism
   - (iv) DGKE mutation
   - (v) Quinine induced
2. Clinical associations
   - (i) HIV
   - (ii) Malignancy, cancer chemotherapy, ionizing radiation
   - (iii) Transplantation, Immunosuppressant use
   - (iv) Pregnancy: HELLP syndrome
   - (v) Autoimmune disease, collagen disease
   - (vi) Others

Comments

As described in Sect. 5.1, aHUS has several etiologies that can affect presentation, management and outcome. Supportive care including dialysis and various type of intensive care to control patient's general conditions is important, as is the case for STEC-associated HUS. Specific therapy is needed for various etiologies. Therefore, we have described the importance of supportive care for treatment of patients with aHUS in the opening statement of this guideline.

1. Pneumococci-associated aHUS

Children with pneumococci-associated HUS are usually younger at presentation and show a more severe course than those with STEC-associated HUS. The mortality rate of pneumococci-associated HUS in the acute phase has been reported to be 12.5 % [c], and 26 % [l]. It has been reported that 10.1 % [c], with 8 % of patients develop end-stage renal disease [l]. These rates are between two to three times higher than those for STEC-associated HUS. As for the pathophysiology of pneumococci-associated HUS, it has been proposed that neuraminidase, produced by pneumococci, cleaves *N*-acetyl neuraminic acid from the cell surface of erythrocytes, platelets, and glomerular endothelial cells, exposing the Thomsen–Friedenreich antigen. The latter is identified by a natural IgM antibody as a normal plasma constituent that initiates the cascade of events leading to HUS. Transfusion of plasma products containing anti-Thomsen–Friedenreich IgM antibodies further accelerates hemolysis, and such cases have been documented [2, 3]. The reported morbidity rate of CKD or end-stage renal disease is significantly lower in patients treated with washed blood products than in those treated with unwashed products [1]. These circumstances dictate that plasma therapy, including plasma infusion and plasma exchange with fresh frozen plasma, should not be performed in patients with pneumococci-associated HUS. Washed blood products should be used for blood transfusion and filler in the dialysis circuit for infant dialysis.

2. aHUS associated with complement dysregulation and other abnormalities

The guideline recommends that daily plasma therapy, including plasma exchange and plasma infusion, should be started at the point of diagnosis of aHUS (excluding cobalamine metabolism disorder for which vitamin B12 supplementation is the established therapy) [d, h]. Plasma exchange is commonly undertaken daily using 1.5–2 plasma volume per session, employing fresh frozen plasma. Plasma exchange is more efficient than plasma infusion, as the former supplies a large amount of normal complement regulatory protein, avoids any risk of volume overload, and removes fluid-phase causative agents (such as abnormal complement regulatory proteins, anti-CFH antibodies, inflammatory cytokines, and other triggers of platelet hyperaggregability) [f, h]. The results of a case series study suggested that the response to short-term plasma therapy varies according to genotype [j, k, 4, 5]. Patients with CFH mutations have the poorest prognosis. On the other hand, patients with MCP mutations have the best short-term prognosis, with 90 % of such patients reported to survive and remain dialysis-free in the long term [4]. Therefore, in patients with MCP mutations, plasma therapy does not affect outcome. This is consistent with the fact that MCP is not a circulating complement regulatory protein.

In the patients with mutations in genes for complement proteins and their regulators, the outcome of kidney transplantation is poor; overall risk of aHUS recurrence after kidney transplantation is about 50 %, and the risk of graft loss occurs in 80–90 % of patients with recurrence [l–n, 6]. The outcomes of kidney transplantation vary according to the type of mutated gene, being poor inpatients with CFH, complement factor I (CFI), C3 mutations. In contrast, kidney graft outcome is reportedly favorable, and disease recurrence rates are low in patients with MCP mutations, due to the fact that MCP is a transmembrane protein and that kidney grafts show normal expression of MCP [6]. The efficacies and benefits of plasma therapy in the perioperative period have been reported in some case series, with the purpose of preventing aHUS recurrence after kidney transplantation [l–n]. Therefore, preventive perioperative plasma therapy is recommended when