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Editorial Comment

Management of Shiga toxin-associated *Escherichia coli*-induced haemolytic uraemic syndrome: randomized clinical trials are needed

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The production of Shiga toxin by a bacterial strain is necessary for induction of enteropathogenic haemolytic uraemic syndrome (HUS). Only strains that produce the toxin are associated with HUS. These include enterohaemorrhagic *Escherichia coli*, *Shigella dysenteriae* and rarely *Citrobacter freundii* [1, 2]. All strains associated with haemorrhagic colitis and HUS are Gram-negative, thus producing lipopolysaccharide (LPS), which may also play a role in the pathogenesis of disease. The recent large outbreak of *E. coli* O104:H4, a strain with combined virulence factors characteristic of enteroaggregative *E. coli* as well as enterohaemorrhagic *E. coli*, provides additional evidence that, regardless of the bacterial background, acquirement of the bacteriophage-encoded gene for Shiga toxin 2 enhances the virulence of the strain causing haemorrhagic colitis and HUS. In this issue of NDT, Kielstein *et al.* [3] present a comprehensive summary of the outcome of this outbreak and the treatment options offered in 491 patients. Appropriate management of Shiga toxin-induced disease is contingent on an understanding of the pathogenesis of the disease as depicted in Figure 1. After ingestion, the bacteria may bind to the terminal ileum and follicle-associated epithelium of Peyer's patches [4]. Colonization is further enhanced by quorum sensing, a form of communication with other strains in the intestinal microflora, as well as induction by the host hormonal response including epinephrine and norepinephrine, which would presumably be secreted during haemorrhagic colitis [5]. These signals enhance bacterial colonization and the release of virulence factors. There is no evidence of bacteraemia during infection with Shiga toxin-producing *E. coli.* Following intestinal colonization, the disease is mediated by the systemic spread of bacterial virulence factors, of which Shiga toxin is of major importance.

Shiga toxin binds to the globotriaosylceramide Gb3 receptor on Paneth cells [6] and is translocated through the intestinal epithelium [7]. The toxin has been shown to induce dysentery [8] and intestinal apoptosis [9]. The inflammatory host response in the intestine is important for the clearance of bacteria from the gut. A reduced intestinal response increases the bacterial burden and thus enables more circulating bacterial virulence factors, as demonstrated in mice [10].

Bacterial virulence factors gain access to the circulation after passing through the intestinal mucosa and damaging the intestinal endothelium. During HUS, Shiga toxin, as well as LPS, circulate bound to platelets, monocytes and neutrophils as well as aggregates between these blood cells [11–14]. Both Shiga toxin and O157LPS induce the formation of platelet and monocyte microparticles bearing tissue factor and complement [14, 15]. These circulating microparticles were also detected during the recent *E. coli* O104 outbreak [16]. Thus, blood cell activation may contribute to the thrombotic process.

It is unknown whether and how circulatory toxin is transferred from blood cells to Gb3-expressing target organ cells *in vivo*. It has been suggested that this transfer of toxin may be related to a higher affinity for the glycolipid receptor on endothelial cells in the kidney [17, 18]. Shiga toxin induces glomerular endothelial cell injury and secretion of tissue factor as well as chemokines promoting leukocyte adhesion to endothelial cells [19, 20]. This scenario enables thrombus formation and release of leukocyte proteases and cytokines. In addition, Shiga toxin induces injury and apoptosis of renal cortical tubular cells [21], and the combined effect on glomeruli and tubuli will lead to destruction of the nephron. The brain is also a target organ in Shiga-toxin-induced disease and studies have shown that the toxin injures endothelial cells as well as neurons [22].

To date, there is no specific treatment for Shiga-toxin-mediated infection. In light of the known pathogenesis of disease, various management approaches can be selected (Table 1). In the study by Kielstein *et al.* [3], three treatment strategies were compared, best supportive care, and plasma exchange with, or without, eculizumab.

Best supportive care would include volume replacement, parenteral nutrition and dialysis. Volume expansion with isotonic solutions administered during the

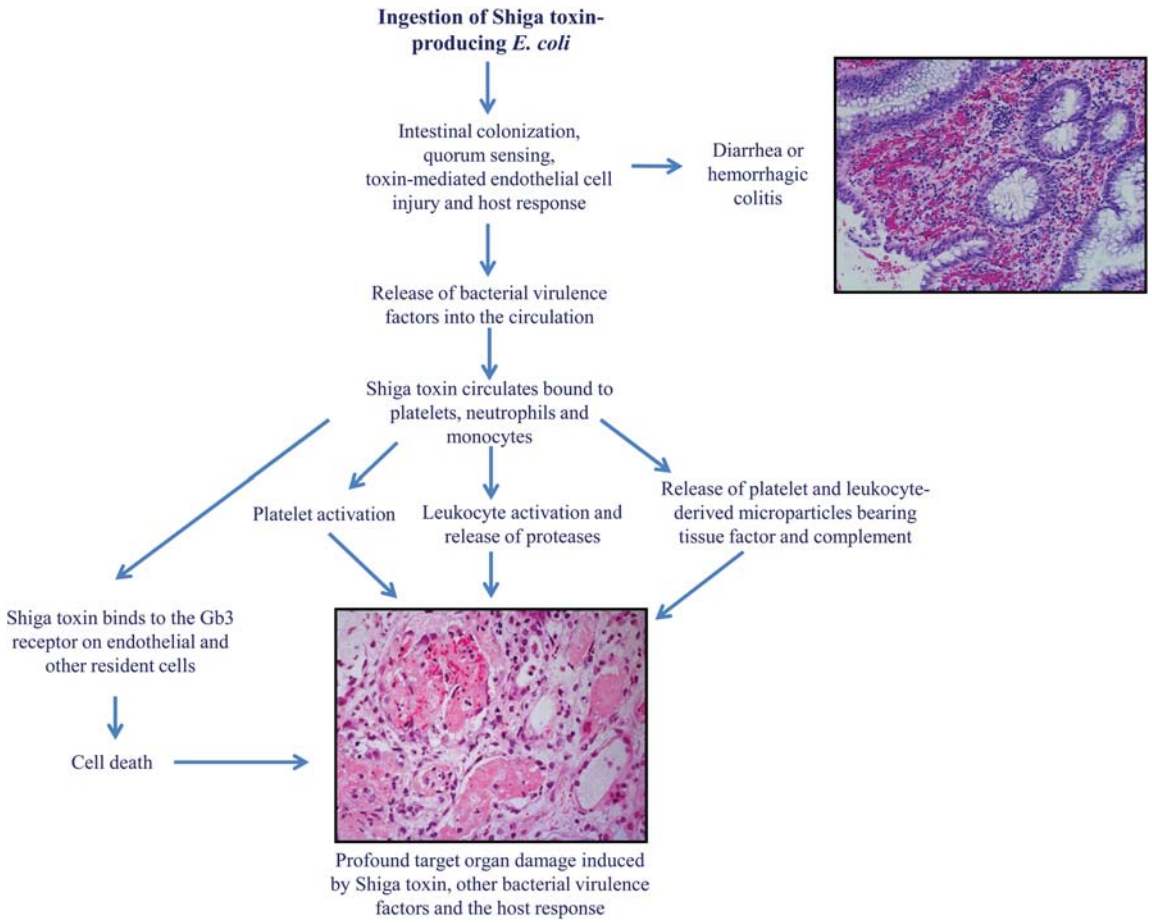

Fig. 1. Schematic presentation of the pathogenesis of Shiga toxin-induced disease.

first 4 days of infection and pre-HUS was found to decrease the rate of oligo-anuria and the need for dialysis [23]. Rehydration may mitigate toxin-mediated tubular injury as well as the formation of microvascular clots. Likewise, diuretics plus rehydration could potentially have a beneficial effect on kidney perfusion, as long as there is residual tubular function, although this is debatable [24]. The need for dialysis, and time at which to commence, is comparable with other conditions associated with acute renal failure [25]. In children, peritoneal dialysis is usually the preferred modality of treatment, whereas haemodialysis was the chosen modality for most adult patients during the O104 outbreak. Of note, most children with HUS during the O104 outbreak received supportive care only (67/90, 74%) and the short-term outcome was similar to previous outbreaks of Shiga toxin-producing *E. coli* [26].

The issue of antibiotic treatment to eradicate the bacterial strain has previously been addressed, most recently in a multi-centre investigation of children with *E. coli* O157:H7 [27]. Regardless of the antibiotic given, children treated with antibiotics during the diarrhoeal phase more frequently developed HUS than those who did not. This has been attributed to toxin release from bacteria during the early phase of treatment, due to bacteriophage lysogenesis. In mice, the bacterial burden, and presumed toxin release, could be correlated to the severity of symptoms [9, 10]. Thus, the presence of a viable bacterium in the intestine, continuously releasing toxin, could affect the course of disease. Whether antibiotic treatment during HUS has a beneficial or deleterious effect remains to be evaluated. The *E. coli* O104 strain associated with the recent outbreak had an extended-spectrum beta lactamase phenotype. The German Society for Infectious Diseases recommended antibiotic treatment under certain circumstances, such as invasive intestinal infection, eradication of meningococci when eculizumab was used, or for reduction of the intestinal bacterial count if colonization was persistent (www.ehec-register.de). Patients who received antibiotics during the outbreak exhibited fewer seizures, lower morbidity, required no abdominal surgery and excreted the *E. coli* strain for a shorter time [28]; thus, antibiotic treatment during ongoing HUS may be beneficial.

Plasma exchange has been attempted in patients with HUS. The rationale would be to remove bacterial toxin, prothrombotic factors, inflammatory mediators and/or blood cell-derived microparticles and to replenish coagulation and complement factors. The amounts of free Shiga toxin in the circulation are negligible [12] but toxin may

Table 1. Treatment options for Shiga toxin-producing *E. coli* infection

Treatment	Rationale	Comment	Outcome	References
Volume expansion	Kidney perfusion during the first days after onset of diarrhoea	Isotonic solutions preferred for volume replacement	Treatment pre-HUS prevents oligo-anuria	[23, 40]
Diuretics	Kidney perfusion during acute renal failure		A single study showed a beneficial effect on diuresis	[41]
Blood transfusion	Packed red blood cells for severe anaemia Hb<60 g/L[a]	Platelet transfusions should be reserved for patients undergoing surgery or during major bleeding episodes	Platelet transfusions may worsen the clinical course	[42, 43]
Dialysis	Management of hypervolaemia, hyperkalaemia, severe acidosis and uraemia	Choice of dialysis modality is dependent on the centre and if the patient has undergone abdominal surgery. Anti-coagulation should be tightly monitored	Early initiation of dialysis has no proven benefit	[25, 44]
Antibiotics	Antimicrobial effect to reduce the bacterial load	Antibiotic treatment in the pre-HUS stage may increase the risk of developing HUS	The effect of antibiotics after HUS-onset may be beneficial	[27,28,45]
Plasma exchange	Removal of toxin, inflammatory mediators and/or blood cell degradation products	Risk of allergic reactions	No benefit shown in children or adults. Controlled studies required	[25, 26, 28–30, 46]
Immunoadsorption	Removal of a presumptive antibody	STEC-mediated HUS has not been shown to be antibody-mediated	Benefit shown in a single study. Controlled studies required	[47]
Eculizumab	Blockade of the terminal complement pathway	Patients should be vaccinated against meningococci or treated with antibiotics Treatment is costly	Benefit in single paediatric cases. No clear benefit in the German *E. coli* O104 outbreak. Controlled studies required	[3, 26, 28, 33]
Anti-coagulation/anti-thrombosis[b]	Prevention of thrombotic microangiopathy	Risk of bleeding	No benefit	[48–50]
Corticosteroids	Reduction of the inflammatory response		No effect on haematological, neurological, or nephrological parameters	[28, 51]
Potential future treatments				
Monoclonal antibodies to Shiga toxin	Neutralize Stx1 and/or 2	Well tolerated in healthy volunteers	Urtoxazumab was tested in infected children. Phase 2/3 trial of ShigaMabs is ongoing	[25, 52–54]
Gb3 receptor analogues	Bind Stx	Tested in mice	Not yet tested in humans	[55, 56]
Manganese	Blocks intracellular trafficking of Stx	Tested *in vivo* in mice and *in vitro*	Not yet tested in humans	[57]
Vaccines	Directed to: • *E. coli* O157:H7 O-specific polysaccharide • Stx 2 • Stx 2B-1B fusion protein • EspA, intimin and Stx2 • Stx2 A2 and B subunits	Tested in healthy children or in mice	Not yet tested in affected individuals	[58–62]

Stx, Shiga toxin; EspA, *E. coli* secreted protein A.
[a]This recommendation is for children; adults may develop symptomatic anaemia and require transfusion at higher haemoglobin levels.
[b]References refer to trials or cases testing heparin, heparin plus dipyramidole and heparin plus urokinase.

be bound to blood cells or microparticles in the circulation [14, 16]. These microparticles possess tissue factor and complement factors on their surfaces [14, 15]. If they are not phagocytosed they may be prothrombotic. Thus, their elimination could theoretically be beneficial. A study carried out in 47 patients during the recent *E. coli* O104 outbreak showed that leukocyte-derived microparticles were removed by plasma exchange [16]. There is, however, insufficient evidence that plasma exchange is beneficial in Shiga toxin-mediated disease. In children, plasma does not provide an additive effect to supportive care alone [26, 29], but there is anecdotal evidence for a certain effect in adults [30, 31], although these studies were not controlled. During the O104 outbreak most adult patients with HUS were treated with plasma exchange, with or without eculizumab [3, 28]. Patients treated with

plasma presented worse symptoms and signs of disease at onset, compared with those treated with supportive care, which may have affected the degree of residual symptoms and higher creatinine levels at discharge [3]. Although a comparison between best supportive care and therapeutic plasma exchange was not possible, the authors suggested that better general medical care, rather than frequent plasma exchange, may have accounted for the good outcome during this outbreak [3]. A recent study validating the treatment strategies given during the *E. coli* O104 outbreak reported no benefit of plasma exchange [28] and similar conclusions were drawn in a review of interventions in HUS [32].

At the time of the outbreak of *E. coli* O104, a publication in *New England Journal of Medicine* described the use of eculizumab, a monoclonal anti-C5 antibody (Soliris, Alexion), in three paediatric cases of HUS associated with other strains of Shiga toxin-producing *E. coli* [33]. This prompted the German Society of Nephrology to recommend the use of eculizumab for the sickest patients during this outbreak. The patients suitable for this treatment were defined as having an infection with Shiga toxin-producing *E. coli* or bloody diarrhoea, as well as neurological symptoms and/or acute kidney injury stadium III and/or venous or arterial thromboembolic events. In the paper by Kielstein *et al.* [3], 193/491 patients were treated with eculizumab in addition to plasma exchange. Eculizumab is approved for treatment of the complement-mediated diseases paroxysmal nocturnal haemoglobinuria and atypical HUS. The rationale for its use in enteropathogenic HUS is that complement activation via the alternative pathway occurs during this form of HUS [15, 34, 35] although this appears to be a secondary phenomenon and not necessarily related to the degree of renal injury [35]. The results presented thus far from the *E. coli* O104 outbreak do not support the use of eculizumab in adults or children with HUS [3, 26, 28]. Treatment did not affect levels of platelet-derived microparticles but seemed to increase the numbers of circulating dead leukocytes [16]. Randomized clinical trials are needed to determine whether certain patients with severe symptoms may benefit from this treatment. Treatment with an antibody that blocks the terminal complement pathway may have deleterious effects and prolong bacterial survival in the gut. The complement system is active in the colon [36, 37] and blocking its function would be expected to increase the bacterial burden. As a consequence, patients treated with eculizumab may have prolonged toxin release if not treated with antibiotics to eradicate the strain. It is thus important that future studies also address the issue of bacterial survival.

During the *E. coli* O104 outbreak, 3842 individuals were infected in Germany, 855 developed HUS and 54 patients died (1.4% mortality) [28, 38]. The study presented in this issue [3] also included patients treated in Sweden and the Netherlands in which a total mortality rate of 4.1% was described for 491 HUS patients. Importantly, not all cases of death occurred in HUS patients. The authors provide the causes of death in HUS patients showing that many patients died of severe complications not necessarily related to the degree of renal failure, as 7/20 patients who died were not on dialysis, and that mortality occurred in older patients. The mortality rate raises the question whether this strain is hyper-virulent in comparison with other Shiga toxin-producing *E. coli* strains. Although 26% of affected children developed neurological symptoms, in similarity to previous outbreaks [26], the percentage of neurological symptoms in affected adults was higher (69%) and the neurological symptoms occurred in a biphasic manner after signs and symptoms of HUS were improving [39], thus presenting an unexpected course of disease. This emergent strain of Shiga toxin-producing *E. coli* differs in its clinical presentation in adults. Although the short-term outcome in children was similar to previous outbreaks [26], the course of disease in adults may require management strategies different from those used over the years in children. The efficacy of available and future treatments should therefore be evaluated by randomized clinical trails in order to establish which treatment strategies are most beneficial to patients.

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Conflict of interest statement. D.K. was the national coordinator in Sweden of the multi-centre international trial of eculizumab (Alexion Pharmaceuticals) in patients with atypical haemolytic uraemic syndrome (2010).

(See related article by Kielstein *et al.* Best supportive care and therapeutic plasma exchange with or without eculizumab in Shiga-toxin-producing *E. coli* O104:H4 induced haemolytic–uraemic syndrome: an analysis of the German STEC-HUS registry. *Nephrol Dial Transplant* 2012; 27: 3807–3815.)

References

1. Taylor CM. Enterohaemorrhagic *Escherichia coli* and *Shigella dysenteriae* type 1-induced haemolytic uraemic syndrome. *Pediatr Nephrol* 2008; 23: 1425–1431
2. Tschape H, Prager R, Streckel W *et al*. Verotoxinogenic *Citrobacter freundii* associated with severe gastroenteritis and cases of haemolytic uraemic syndrome in a nursery school: green butter as the infection source. *Epidemiol Infect* 1995; 114: 441–450
3. Kielstein JT, Beutel G, Fleig S *et al*. Best supportive care and therapeutic plasma exchange with or without eculizumab in Shiga toxin-producing *E. coli* O104:H4-induced hemolytic uremic syndrome: an analysis of the German STEC-HUS registry. *Nephrol Dial Transplant* 2012; 27: 3807–3815
4. Phillips AD, Navabpour S, Hicks S *et al*. Enterohaemorrhagic *Escherichia coli* O157:H7 target Peyer's patches in humans and cause attaching/effacing lesions in both human and bovine intestine. *Gut* 2000; 47: 377–381
5. Pacheco AR, Sperandio V. Inter-kingdom signaling: chemical language between bacteria and host. *Curr Opin Microbiol* 2009; 12: 192–198
6. Schuller S, Heuschkel R, Torrente F *et al*. Shiga toxin binding in normal and inflamed human intestinal mucosa. *Microbes Infect* 2007; 9: 35–39

7. Hurley BP, Thorpe CM, Acheson DW. Shiga toxin translocation across intestinal epithelial cells is enhanced by neutrophil transmigration. *Infect Immun* 2001; 69: 6148–6155
8. Fontaine A, Arondel J, Sansonetti PJ. Role of Shiga toxin in the pathogenesis of bacillary dysentery, studied by using a Tox-mutant of *Shigella dysenteriae* 1. *Infect Immun* 1988; 56: 3099–3109
9. Békássy ZD, Calderon Toledo C, Leoj G *et al*. Intestinal damage in enterohemorrhagic *Escherichia coli* infection. *Pediatr Nephrol* 2011; 26: 2059–2071
10. Calderon Toledo C, Rogers TJ, Svensson M *et al*. Shiga toxin-mediated disease in MyD88-deficient mice infected with *Escherichia coli* O157:H7. *Am J Pathol* 2008; 173: 1428–1439
11. Te Loo DM, van Hinsbergh VW, van den Heuvel LP *et al*. Detection of verocytotoxin bound to circulating polymorphonuclear leukocytes of patients with hemolytic uremic syndrome. *J Am Soc Nephrol* 2001; 12: 800–806
12. Brigotti M, Tazzari PL, Ravanelli E *et al*. Clinical relevance of Shiga toxin concentrations in the blood of patients with hemolytic uremic syndrome. *Ped Inf Dis J* 2011; 30: 486–490
13. Ståhl AL, Svensson M, Morgelin M *et al*. Lipopolysaccharide from enterohemorrhagic *Escherichia coli* binds to platelets through TLR4 and CD62 and is detected on circulating platelets in patients with hemolytic uremic syndrome. *Blood* 2006; 108: 167–176
14. Ståhl AL, Sartz L, Nelsson A *et al*. Shiga toxin and lipopolysaccharide induce platelet-leukocyte aggregates and tissue factor release, a thrombotic mechanism in hemolytic uremic syndrome. *PLoS ONE* 2009; 4: e6990
15. Ståhl AL, Sartz L, Karpman D. Complement activation on platelet-leukocyte complexes and microparticles in enterohemorrhagic *Escherichia coli*-induced hemolytic uremic syndrome. *Blood* 2011; 117: 5503–5513
16. Ge S, Hertel B, Emden SH *et al*. Microparticle generation and leucocyte death in Shiga toxin-mediated HUS. *Nephrol Dial Transplant* 2012; 27: 2768–2775
17. te Loo DM, Monnens LA, van Der Velden TJ *et al*. Binding and transfer of verocytotoxin by polymorphonuclear leukocytes in hemolytic uremic syndrome. *Blood* 2000; 95: 3396–3402
18. Brigotti M, Tazzari PL, Ravanelli E *et al*. Endothelial damage induced by Shiga toxins delivered by neutrophils during transmigration. *J Leukoc Biol* 2010; 88: 201–210
19. Zoja C, Buelli S, Morigi M. Shiga toxin-associated hemolytic uremic syndrome: pathophysiology of endothelial dysfunction. *Pediatr Nephrol* 25: 2231–2240
20. Nestoridi E, Tsukurov O, Kushak RI *et al*. Shiga toxin enhances functional tissue factor on human glomerular endothelial cells: implications for the pathophysiology of hemolytic uremic syndrome. *J Thromb Haemostas* 2005; 3: 752–762
21. Karpman D, Hakansson A, Perez MT *et al*. Apoptosis of renal cortical cells in the hemolytic-uremic syndrome: *in vivo* and *in vitro* studies. *Infect Immun* 1998; 66: 636–644
22. Obata F, Tohyama K, Bonev AD *et al*. Shiga toxin 2 affects the central nervous system through receptor globotriaosylceramide localized to neurons. *J Infect Dis* 2008; 198: 1398–1406
23. Hickey CA, Beattie TJ, Cowieson J *et al*. Early volume expansion during diarrhea and relative nephroprotection during subsequent hemolytic uremic syndrome. *Arch Pediatr Adolesc Med* 2011; 165: 884–889
24. Loirat C, Saland J, Bitzan M. Management of hemolytic uremic syndrome. *Presse Med* 2012; 41: e115–e135
25. Bitzan M, Schaefer F, Reymond D. Treatment of typical (enteropathic) hemolytic uremic syndrome. *Semin Thrombos Hemostas* 2010; 36: 594–610
26. Loos S, Ahlenstiel T, Kranz B *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
27. Wong CS, Mooney JC, Brandt JR *et al*. Risk factors for the hemolytic uremic syndrome in children infected with *Escherichia coli* O157:H7: a multivariable analysis. *Clin Inf Dis* 2012; 55: 33–41
28. Menne J, Nitschke M, Stingele R *et al*. Validation of treatment strategies for enterohaemorrhagic *Escherichia coli* O104:H4 induced haemolytic uraemic syndrome: case-control study. *Br Med J* 2012; 345: e4565
29. Rizzoni G, Claris-Appiani A, Edefonti A *et al*. Plasma infusion for hemolytic-uremic syndrome in children: results of a multicenter controlled trial. *J Pediatr* 1988; 112: 284–290
30. Nakatani T, Tsuchida K, Yoshimura R *et al*. Plasma exchange therapy for the treatment of *Escherichia coli* O-157 associated hemolytic uremic syndrome. *Int J Mol Med* 2002; 10: 585–588
31. Colic E, Dieperink H, Titlestad K *et al*. Management of an acute outbreak of diarrhoea-associated haemolytic uraemic syndrome with early plasma exchange in adults from southern Denmark: an observational study. *Lancet* 2011; 378: 1089–1093
32. Michael M, Elliott EJ, Ridley GF *et al*. Interventions for haemolytic uraemic syndrome and thrombotic thrombocytopenic purpura. *Cochrane Database Syst Rev* 2009: CD003595
33. Lapeyraque AL, Malina M, Fremeaux-Bacchi V *et al*. Eculizumab in severe Shiga-toxin-associated HUS. *N Engl J Med* 2011; 364: 2561–2563
34. Monnens L, Molenaar J, Lambert PH *et al*. The complement system in hemolytic-uremic syndrome in childhood. *Clin Nephrol* 1980; 13: 168–171
35. Thurman JM, Marians R, Emlen W *et al*. Alternative pathway of complement in children with diarrhea-associated hemolytic uremic syndrome. *Clin J Am Soc Nephrol* 2009; 4: 1920–1924
36. Ueki T, Mizuno M, Uesu T *et al*. Distribution of activated complement, C3b, and its degraded fragments, iC3b/C3dg, in the colonic mucosa of ulcerative colitis. *Clin Exp Immunol* 1996; 104: 286–292
37. Chen G, Yang Y, Gao X *et al*. Blockade of complement activation product C5a activity using specific antibody attenuates intestinal damage in trinitrobenzene sulfonic acid induced model of colitis. *Lab Invest* 2011; 91: 472–483
38. Frank C, Werber D, Cramer JP *et al*. Epidemic profile of Shiga-toxin-producing *Escherichia coli* O104:H4 outbreak in Germany. *N Engl J Med* 2011; 365: 1771–1780
39. German EHEC-HUS Registry. The German 2011 epidemic of Shiga toxin-producing *E. coli*—the nephrological view. *Nephrol Dial Transplant* 2011; 26: 2723–2726
40. Ake JA, Jelacic S, Ciol MA *et al*. Relative nephroprotection during *Escherichia coli* O157:H7 infections: association with intravenous volume expansion. *Pediatrics* 2005; 115: e673–e680
41. Rousseau E, Blais N, O'Regan S. Decreased necessity for dialysis with loop diuretic therapy in hemolytic uremic syndrome. *Clin Nephrol* 1990; 34: 22–25
42. Karpman D. Haemolytic uremic syndrome and thrombotic thrombocytopenic purpura. *Current Paediatr* 2002; 12: 569–574
43. George JN, Vesely SK. Thrombotic thrombocytopenic purpura-hemolytic uremic syndrome: diagnosis and treatment. *Cleve Clin J Med* 2001; 68: 857–858
44. Trachtman H, Cnaan A, Christen E *et al*. Effect of an oral Shiga toxin-binding agent on diarrhea-associated hemolytic uremic syndrome in children: a randomized controlled trial. *J Am Med Assoc* 2003; 290: 1337–1344
45. Wong CS, Jelacic S, Habeeb RL *et al*. The risk of the hemolytic-uremic syndrome after antibiotic treatment of *Escherichia coli* O157:H7 infections. *N Engl J Med* 2000; 342: 1930–1936
46. Dundas S, Murphy J, Soutar RL *et al*. Effectiveness of therapeutic plasma exchange in the 1996 Lanarkshire *Escherichia coli* O157:H7 outbreak. *Lancet* 1999; 354: 1327–1330
47. Greinacher A, Friesecke S, Abel P *et al*. Treatment of severe neurological deficits with IgG depletion through immunoadsorption in patients with *Escherichia coli* O104:H4-associated haemolytic

uraemic syndrome: a prospective trial. *Lancet* 2011; 378: 1166–1173
48. Van Damme-Lombaerts R, Proesmans W, Van Damme B *et al*. Heparin plus dipyridamole in childhood hemolytic-uremic syndrome: a prospective, randomized study. *J Pediatr* 1988; 113: 913–918
49. Loirat C, Beaufils F, Sonsino E *et al*. Treatment of childhood hemolytic-uremic syndrome with urokinase. Cooperative controlled trial. *Arch Fr Pediatr* 1984; 41: 15–19
50. Sorrenti LY, Lewy PR. The hemolytic-uremic syndrome: experience at a center in the Midwest. *Am J Dis Child* 1978; 132: 59–62
51. Perez N, Spizzirri F, Rahman R *et al*. Steroids in the hemolytic uremic syndrome. *Pediatr Nephrol* 1998; 12: 101–104
52. Bitzan M, Poole R, Mehran M *et al*. Safety and pharmacokinetics of chimeric anti-Shiga toxin 1 and anti-Shiga toxin 2 monoclonal antibodies in healthy volunteers. *Antimicrob Agents Chemother* 2009; 53: 3081–3087
53. Lopez EL, Contrini MM, Glatstein E *et al*. Safety and pharmacokinetics of urtoxazumab, a humanized monoclonal antibody, against Shiga-like toxin 2 in healthy adults and in pediatric patients infected with Shiga-like toxin-producing *Escherichia coli*. *Antimicrob Agents Chemother* 2010; 54: 239–243
54. Dowling TC, Chavaillaz PA, Young DG *et al*. Phase 1 safety and pharmacokinetic study of chimeric murine-human monoclonal antibody c alpha Stx2 administered intravenously to healthy adult volunteers. *Antimicrob Agents Chemother* 2005; 49: 1808–1812
55. Mulvey GL, Marcato P, Kitov PI *et al*. Assessment in mice of the therapeutic potential of tailored, multivalent Shiga toxin carbohydrate ligands. *J Infect Dis* 2003; 187: 640–649
56. Watanabe M, Matsuoka K, Kita E *et al*. Oral therapeutic agents with highly clustered globotriose for treatment of Shiga toxigenic *Escherichia coli* infections. *J Infect Dis* 2004; 189: 360–368
57. Mukhopadhyay S, Linstedt AD. Manganese blocks intracellular trafficking of Shiga toxin and protects against Shiga toxicosis. *Science* 2012; 335: 332–335
58. Ahmed A, Li J, Shiloach Y *et al*. Safety and immunogenicity of *Escherichia coli* O157 O-specific polysaccharide conjugate vaccine in 2–5-year-old children. *J Infect Dis* 2006; 193: 515–521
59. Wen SX, Teel LD, Judge NA *et al*. A plant-based oral vaccine to protect against systemic intoxication by Shiga toxin type 2. *Proc Natl Acad Sci* 2006; 103: 7082–7087
60. Gao X, Cai K, Shi J *et al*. Immunogenicity of a novel Stx2B-Stx1B fusion protein in a mice model of Enterohemorrhagic *Escherichia coli* O157:H7 infection. *Vaccine* 2009; 27: 2070–2076
61. Gu J, Liu Y, Yu S *et al*. Enterohemorrhagic *Escherichia coli* trivalent recombinant vaccine containing EspA, intimin and Stx2 induces strong humoral immune response and confers protection in mice. *Microbes Infect* 2009; 11: 835–841
62. Bentancor LV, Bilen M, Brando RJ *et al*. A DNA vaccine encoding the enterohemorragic *Escherichia coli* Shiga-like toxin 2 A2 and B subunits confers protective immunity to Shiga toxin challenge in the murine model. *Clin Vaccine Immunol* 2009; 16: 712–718

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Pulmonary hypertension in dialysis patients: a prevalent, risky but still uncharacterized disorder

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Pulmonary hypertension (PH) is a complex hemodynamic alteration which may result from disparate causes. In 1973 at a conference endorsed by the World Health Organization (WHO), a classification based on two categories only (primary and secondary PH) was proposed [1]. In 2001, a new classification establishing five categories of PH supplanted the first classification [2]. Since then, minor modifications were made and the last (2008) WHO classification maintains five diagnostic categories (Table 1) [3]. In the 2008 classification by the WHO and in more recent guidelines by the European Society of Cardiology (ESC) [4], for the first time attention was given to PH in dialysis patients which was classified in the fifth category, i.e. in a limbo category gathering various forms of PH 'with unclear or multifactorial etiology'. At that time only one survey in dialysis patients was available [5] and this report showed an unexpectedly high prevalence of PH which was mainly attributed to high cardiac output secondary to the presence of arterio-venous fistula [6], anemia and/or fluid overload and to left ventricular (LV) disorders. During the last 5 years, PH in patients with kidney diseases has attracted increasing attention and over 100 original or review articles dealing with PH in dialysis patients or in predialysis chronic kidney disease (CKD) and in transplant patients are now deposited in PubMed. In this issue of NDT, Rajiv Agarwal reports on the largest study performed so far in