Haemolytic uraemic syndrome associated with a family cluster of enterohaemorrhagic *Escherichia coli*

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Introduction

In early 1995 an outbreak of 23 cases of haemolytic uraemic syndrome (HUS) occurred in children (ranging from four months to 12 years of age) in South Australia.[1] Twenty of the cases were managed in a tertiary paediatric hospital in Adelaide, where 18 (90%) required dialysis.[2] A 4-year-old died and 12 months after discharge 5 of the surviving children still had significantly impaired renal function.[2]

In Australia HUS is usually caused by a subgroup of Shiga toxin-producing *Escherichia coli* known as enterohaemorrhagic *E. coli* (EHEC). The Shiga toxins cause cell damage and trigger an inflammatory process which initiates intravascular coagulation resulting in microthrombi forming in small blood vessels in the gut and kidney.[3] The natural reservoir of EHEC is the gut of animals, particularly cattle and sheep. Hence HUS can be caused by contact with animal faeces, either directly or via contaminated, inadequately cooked food, particularly meat and dairy products. Most of the cases in the 1995 outbreak of HUS in South Australia had consumed (in the week before the onset of illness) an uncooked fermented sausage manufactured in Adelaide.[1] Subsequent molecular studies revealed an identical EHEC in both faeces of the cases and samples of the sausage.[4]

Because EHEC infection, and therefore HUS, can be foodborne, it is of considerable public health concern. Following the South Australian outbreak, HUS became a notifiable disease in Queensland in mid-1996, and EHEC in mid-2001. However, a recent case of HUS in north Queensland has identified several shortcomings in the management and investigation of HUS and EHEC infections; some of these shortcomings were also identified in a previous cluster of HUS cases that occurred in north Queensland in 2004.[5]

The HUS case and subsequent investigations

In early January 2007, the Tropical Population Health Network (TPHN) was notified by an infection control practitioner that a 14-month-old Caucasian girl had been hospitalised the previous day with HUS; she had become unwell four days before being hospitalised. *Salmonella* Virchow was isolated from a diarrhoeal stool sample collected two days prior to her being hospitalised and from a stool sample collected on the day of admission.

The attending physician initially believed that the *Salmonella* infection was the cause of the HUS, and this led to problems in getting the (diarrhoeal) samples to the Queensland Health Scientific Services reference laboratory for screening for Shiga toxin (stx1 and stx2) gene (and therefore for EHEC). The initial sample was not forwarded, and the second sample was not forwarded frozen to the laboratory. When the latter sample was eventually screened stx genes were not detected. Therefore EHEC was never detected in the HUS case.

The child's parents were interviewed using the relevant OzFoodNet questionnaire; this did not reveal any suspect food items in the child's diet. However, it did reveal that the child and her two siblings had visited several commercial animal sanctuaries during the exposure period. At two of these the children had had direct contact with marsupials (particularly kangaroos and koalas) and apparently also with faeces from these animals. The parents stated that the children had no contact with other mammals at these sanctuaries, and no apparent contact with any bovine animals during the exposure period.

The child's two siblings attended a local child-care centre. Even though they were apparently asymptomatic, stool samples were collected (in mid-January) from both and the parents were requested by TPHN to keep them out of child care until the results of the stool tests were known.[6,7] The child's twin sibling's stool was positive for the stx2 (but not the stx1) gene and the eaeA gene (which encodes a virulence factor: intimin) upon screening, and was culture positive for *E. coli* O55:H80, *S.* Aberdeen and *S.* Chailey. The child's 3-year-old brother's stool was also positive for the stx2 (but not the stx1) gene and the eaeA gene upon screening, and was culture positive for *E. coli* O55:HR. ('R' indicates that the organism had become rough in sub-cultures; once an EHEC becomes rough, the H antigen cannot be typed.) Both parents then had stool samples collected, but neither had evidence of EHEC upon screening.

The two siblings were voluntarily excluded from child-care until they were clear of the Shiga toxin in weekly stool samples. The 3-year-old and the twin sibling were able to return to child-care (after having two successive stool samples collected at least 48 hours apart clear of any evidence of Shiga toxin or EHEC[7]) 3.5 and 4.5 weeks, respectively, after having been first identified as being infected with EHEC. This delay created considerable difficulties for the parents, and repeated explanations of the importance of their exclusion from child-care were necessary.

The two siblings (and presumably the case) were infected with Shiga toxin-producing *E. coli* (all presumably with O55:H80), and the twins were infected with three different *Salmonella* serovars. This array of pathogens supported the hypothesis, as suggested from the parent interview, that the EHEC was acquired via animal contact rather than via a particular food item. For this reason, an assessment of the facilities and signs at the two animal sanctuaries was undertaken (about 2.5 weeks after the onset of the HUS) by environmental health officers. The public was encouraged to handle animals at both sanctuaries but there were no signs recommending hand washing after handling the animals at either sanctuary. At one sanctuary there were no hand washing facilities near the animal-handling areas. There were several food outlets in close proximity to the animal facilities.

It appeared that the management of the sanctuaries had little understanding of the potential infectious hazards associated with such facilities, and were uncertain of their responsibilities to minimise the risk of such hazards. Indeed, there are no guidelines in Queensland on how to minimise these risks for managers of commercial facilities that encourage the public to handle animals.

There was no evidence of Shiga toxin in faecal samples from koalas and kangaroos at either of the facilities, however, the samples were collected about a month after the onset of the HUS.

Discussion

This report describes three siblings infected with a Shiga toxin-producing EHEC; two who remained asymptomatic (presumably both with *E. coli* O55:H80), and their sibling (presumably infected with the same EHEC) who developed HUS. Both the sxt2 and eaeA genes were detected in this EHEC; this combination of genes appears to be an important predictor of HUS.[8]

This is the second cluster of EHEC with HUS in north Queensland in three years.[5] The two clusters have identified several issues of concern (Box).

Salmonella infections do not cause HUS,[9] and the isolation of salmonellae from faecal samples from a HUS patient must be regarded as coincidental to the HUS. Screening for Shiga toxin and other virulence genes was undertaken on the *S.* Virchow isolated from the HUS child to prove this point to the attending physician; none of the genes were detected. Faecal samples from HUS cases must be forwarded promptly to a reference laboratory for screening for EHEC regardless of the isolation of *Salmonella* from the samples. Failure to do this may result in EHEC not being isolated from a case (as happened with the child with HUS in this cluster), and could impede the necessary investigations.

Issues of public health significance revealed by this family cluster of EHEC infections

- HUS is a notifiable disease. As it is a syndrome, notification has to come from clinicians: not only paediatricians but also haematologists, nephrologists, infectious diseases and intensive care physicians need to be aware of their responsibility to notify cases of HUS.
- As soon as HUS is diagnosed, a stool sample should be sent to the relevant reference laboratory for screening tests for Shiga toxins and EHEC.
- *Salmonella* infections do not cause HUS. Isolation of *Salmonella* from the stool of an HUS case must be considered as coincidental to the HUS.
- Stool samples being submitted for investigation for Shiga toxin and EHEC must be frozen soon after collection, and transported frozen to the reference laboratory.
- Children less than 5 years of age who attend child-care, and who are household contacts of a case of symptomatic EHEC infection, should be screened for EHEC even if they are asymptomatic. They should be excluded from child-care until two stool samples, collected at least 48 hours apart, are shown to be clear of the EHEC.
- Infectious disease hazards are associated with contact with animals in public facilities – 'petting zoos' – and the management of such facilities need to take measures to reduce the risk of these hazards.
- Guidelines on how these infectious hazards can be minimised need to be formally endorsed by the relevant agencies so that the management of petting zoos and other similar facilities can be made aware of their responsibilities.

The infecting dose of HUS is very low, and person-to-person transmission of EHEC is well documented, with transmission occurring among young children within families and in child care facilities.[10] For this reason it is essential to screen the young siblings of EHEC HUS cases for the organism even if they are asymptomatic, and older siblings (and other close contacts) if they have any relevant symptoms.[5] These individuals should be excluded from child care (or any workplace of concern) while the screening takes place, and may need to be further excluded should the screening indicate an EHEC infection.[6,7]

There does not appear to be any published information as to whether marsupials act as reservoirs of EHEC. However, it is well recognised that macropods (kangaroos, wallabies) can be infected with salmonellae,[11] and an outbreak of human salmonellosis associated with contact with wallabies in a petting zoo has been reported from the United States of America.[12] Several other zoonotic (mostly enteric) infections have occurred following handling animals in petting zoos.[12,13]

It is important that guidelines on how to minimise the risk of transmission of infections through handling animals be made readily available to those facilities that encourage the public to handle animals on-site. These guidelines should include educating the public; appropriate signage; providing hand washing facilities; ensuring adequate supervision of children; discouraging eating in animal contact areas; ensuring sick animals are not handled by the public; providing appropriate cleaning and infection of the animal holding area and ensuring the safe disposal of animal faeces.[13] Such guidelines are available in several countries,[14,15] and in South Australia;[16] these guidelines are being used as templates for the drafting, currently in progress, of petting zoo guidelines for use in Queensland.

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