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Possible Cause of Liver Failure in Patient with Dengue Shock Syndrome

To the Editor: We report a rare hepatic ultrasonograph finding for a patient with liver failure associated with dengue virus (DENV) infection. This finding might shed light on the pathogenesis of liver involvement in this disease.

In March 2006, a 10-year-old previously healthy boy was hospitalized for a 3-day history of fever, headache, and nausea/vomiting. Fever subsided on the day of admission, but the patient was in shock (blood pressure 80/40 mm Hg) and had gastrointestinal bleeding and hematuria. Physical examination showed an obese, confused patient with generalized petechiae and hepatomegaly. The initial diagnosis was dengue shock syndrome (DSS). The patient was intubated and received intravenous fluid infusion, packed red blood cells, ceftriaxone, sodium bicarbonate, and ranitidine before being transferred to King Chulalongkorn Memorial Hospital in Bangkok. The patient's blood pressure increased to 130/90 mm Hg after the initial fluid resuscitation (28 mL/kg free flow), and systolic pressure remained at ≈130 mm Hg until transfer.

Laboratory examinations found 14,930 leukocytes/mm^3, hemoglobin 16.4 g/dL, hematocrit 48.2%, platelet 18,000/mm^3, blood urea nitrogen 33 mg/dL, creatinine 1 mg/dL, sodium 128 mEq/L, potassium 6.2 mEq/L, chloride 91 mEq/L, total CO_2 5 mEq/L, total bilirubin 6.9 mg/dL, direct bilirubin 3.9 mg/dL, aspartate transaminase 3,507 IU/L, alanine transaminase 2,775 IU/L, prothrombin time 43 seconds (international normalized ratio 3.4), and partial thromboplastin time 93.5 s (control 28.7 s). Blood and urine cultures showed negative results. Serum was positive for IgM against DENV. Unfortunately, we did not investigate other viral causes of liver failure.

DSS with liver failure was diagnosed and treated with intravenous fluid, sodium bicarbonate, omeprazole, fresh frozen plasma, platelet transfusion, vitamin K, and recombinant factor VIIa concentrate (NovoSeven; Novo Nordisk, Bagsvaerd, Denmark). Despite stable blood pressure over the next 6 days, liver enzymes continued to rise with progressive jaundice (online Technical Appendix, wwwnc.cdc.gov/EID/article/19/7/12-1820-Techapp1.pdf). Hepatic ultrasonograph on the second

day after admission showed totally reversed direction of portal venous blood flow away from the liver (Figure, panel A), becoming bidirectional on the following day and, finally, reverting to normal direction (although with low velocity) 3 days later (Figure, panel B). Despite improved hemodynamic status, progressive encephalopathy and gastrointestinal bleeding developed and were unresponsive to treatment. Six days later, the patient died of pulmonary hemorrhage and progressive respiratory failure.

DENV infection is one of the most prevalent emerging infectious diseases affecting children and one of the leading causes of liver failure in tropical countries (*1*,*2*). Although liver involvement in patients with dengue hemorrhagic fever is well known, the mechanism for DENV-induced liver injury is still a mystery. Liver autopsy specimens of terminal DSS patients generally showed massive or focal necrosis with little or no recruitment of polymorphonuclear cells or lymphocytes (*3*,*4*). Ultrasonograph images from patients with liver failure caused by acetaminophen poisoning or hepatitis B indicate increased portal vein flow and normal flow velocity to the damaged liver (*5*). Decreased portal vein flow velocity and reversal of the flow direction is seen in the terminal stage of hepatic cirrhosis and a few other conditions such as hepatic sinusoidal obstruction (hepatic veno-occlusive disease), arterioportal fistula, extrahepatic portal vein thrombosis, and hepatic venous outflow obstruction (*6*). This finding is unusual in other instances of toxin- or virus-induced liver failure and might contribute to the understanding of the mechanism of liver involvement in patients with DENV infection.

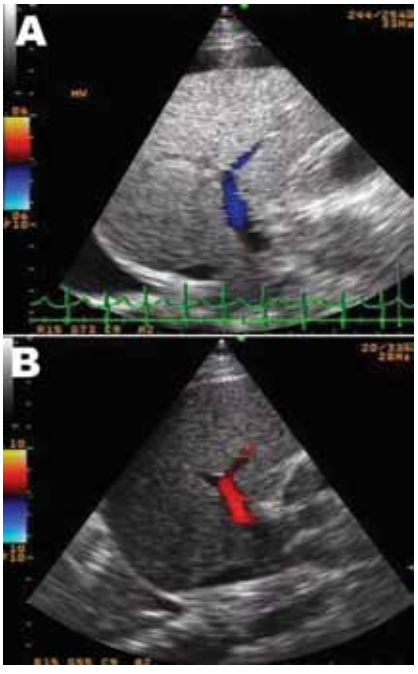

Figure. Ultrasonograph with Doppler image of the liver of a 10-year-old boy with liver failure associated with dengue virus infection. A) Day 2 of hospitalization, showing reversed direction of blood flow in the right branch of the portal vein (hepatofugal flow). There was diffuse increased liver parenchymal echo, swelling of the gallbladder wall, and right pleural effusion. B) Day 5 of hospitalization, showing returning normal direction of portal venous flow (hepatopetal flow). Liver parenchymal echo changed to normal. Pleural fluid and swelling of the gallbladder wall also disappeared.

We previously reported increased portal vein congestion during the toxic stage of DENV infection (*7*). At defervescence, the portal vein was dilated and blood flow velocity was decreased. This finding is usually observed for patients with high resistance in the hepatic sinusoidal capillary network, such as those with liver cirrhosis, and is correlated with the degree of portal venous hypertension (*8*). We postulate that DENV infection of the liver might affect the sinusoidal endothelial or Kupffer cells in a way that causes obstruction to the hepatic sinusoidal capillary lumen resulting in decreased portal venous blood velocity and flow to the liver and, when severe, shunting of portal blood away from the liver (hepatofugal flow). Because portal venous blood comprises 75% of total hepatic blood (*6*), this condition coupled with decreased hepatic arterial blood flow as a consequence of shock might have led to severe and irreversible liver damage in this patient. This hypothesis can be further supported by a pathology study of the skin in patients with DENV infection, which showed endothelial swelling and extrusion of its plasma membrane into the capillary lumen, resulting in narrowing of the capillary lumen (*9*). Of note are the similarities between clinical findings in patients with DENV infection and sinusoidal obstruction syndrome such as hepatomegaly, ascites, right pleural effusion, swelling of the gall bladder wall, and decreased velocity or reversed direction of portal blood flow (*10*).

In conclusion, we report a case of liver failure from DENV infection with reversal of portal venous blood flow. We postulate that hepatic sinusoidal obstruction coupled with shock might be the underlying mechanism of liver failure in this disease.

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Publication of this case report was approved by the ethic committee, Faculty of Medicine, Chulalongkorn University.

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Spotted Fever Group Rickettsiae in Questing Ticks, Central Spain

To the Editor: The number of spotted fever group (SFG) rickettsiae that cause diseases in humans is rapidly increasing (*1*,*2*); infections have been described in ticks and humans in Spain (*3*,*4*). However, in Castilla-La Mancha, central Spain, where recreational parks and hunting estates are abundant and humans may be exposed to infected ticks, information on such infections is not available. Therefore, it is worthwhile to characterize *Rickettsia* spp. found in this area for epidemiologic studies and proper diagnosis of possible rickettsial diseases.

In this study, we obtained 148 questing adult ticks, representing the most abundant species in the area: 12 *Dermacentor marginatus*, 26 *Rhipicephalus bursa*, 41 *Rh. sanguineus*, 15 *Rh. turanicus*, 8 *Rh. pusillus,* 2 *Haemaphysalis punctata*, 11 *Hyalomma lusitanicum,* and 33 *Hyalomma marginatum* (*5*). The ticks were collected from the vegetation at natural sites surveyed in Castilla-La Mancha by blanket dragging with a cotton flannelette during fall 2009 and spring–summer 2010 (Figure, panel A) and classified (*5*).

Total DNA was extracted from dissected tick internal organs by using the DNeasy Blood & Tissue Kit (QIAGEN, Düsseldorf, Germany) and used to analyze *Rickettsia* spp. DNA by PCR, cloning, and sequence analysis of the amplicons. At least 3 clones were sequenced for each amplicon. Genes targeted by PCR included fragments of adenosine triphosphate synthase α subunit (*atpA*), heat-shock protein 70 (*dnaK*), outer membrane protein A (*ompA*), outer membrane protein B (*ompB*), citrate synthase (*gltA*), 16S rRNA, *recA*, and initiator protein of DNA replication (*dnaA*) (*6*,*7*). To characterize *Rickettsia* spp., we compared nucleotide sequence identity to reference strains and carried out multilocus analysis using *ompA-ompB* sequences and in silico *Pst*I and *Rsa*I restriction analysis of *ompA* sequences (*7*).

Ticks were first screened by 16S rRNA PCR, and positive samples were analyzed for all targeted genes. The results showed that 27 (18.2%) of the 148 ticks analyzed were positive for *Rickettsia* spp. Of these, 11 were confirmed as *R. massiliae* in *Rh. sanguineus*, *Rh. turanicus*, and *Rh. pusillus*, 3 as *R. raoultii* in *D. marginatus*, 2 as *R. slovaca* in *D. marginatus*, and 2 as *R. sibirica* subsp. *mongolitimonae* in *H. marginatum* and *Rh. pusillus* (Figure, panel B). These species had >99% pairwise nucleotide sequence identity to reference strains *R. massiliae* MTU5 (GenBank accession no. NC_009900), *R. slovaca* 13-B (accession no. NC_016639), and *R. sibirica* subsp. *mongolitimonae* HA-91 (accession no. AHZB00000000) genome sequences for all genes analyzed, and the only *R. raoultii* reported sequences (accession nos. JQ792107, JQ792166, JQ792134, and NR_043755 for *ompB*, *ompA*, *gltA*, and 16S rRNA, respectively). The sequences obtained in this study were deposited in the GenBank under accession nos. KC427998–KC428040.

Multilocus sequence analysis of *ompA-ompB* sequences (Figure, panel B) and in silico *Pst*I and *Rsa*I restriction analysis of *ompA* sequences also confirmed the identity of the *Rickettsia* spp. identified in this study. As previously shown (*7*,*8*), multilocus analysis with *ompA-ompB* sequences was highly informative about the

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Technical Appendix

Technical Appendix Table. Serial clinical and laboratory data for patient with liver failure and dengue shock syndrome.

	Day after onset of fever							
Parameter	3–4	4–5	5–6†	6–7	7–8	8–9†	9–10	10–11
Heart rate	101–130	100	90	100	100	100	110	100→0
BP, mm Hg‡								
Systolic	80–155	81–151	127–150	122–138	115–130	111–127	106–145	99→0
Diastolic	40–93	52–96	65–90	66–92	57–66	45–60	36–68	32→0
Hematocrit	48.2	24.6	30.7	33.6	27.8	26.9	28.7	24.1
ALT	2,775	4,490	4,720	3,098	2,011	1,729	1,725	543
AST	3,507	11,660	>7,000	8,440	4,082	3,099	2,600	968
Bilirubin								
Total	6.9	9.7	12.2	21.6	24.1	34.8	32.1	23.8
Direct	3.9	5.5	6.3	12.1	14.2	18.4	6.0	14.1
PT, INR	3.4	2.4	2.4	NA	2.3	2.3	2.1	NA

*BP, blood pressure; ALT, alanine transaminase; AST, aspartate transaminase; PT, prothrombin time; INR, international normalized ratio); NA, not available.
†Dates when liver ultrasonography (Figure) was performed.
‡1 episode of BP 81–85/52–54 mm Hg occurred for 15 minutes, 12 hours after hospital admission; otherwise, the lowest systolic BP was 108 mm Hg.