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Short Communication

Hepatitis E virus genotype 1f outbreak in Bangladesh, 2018

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Hepatitis E virus (HEV) infection is a significant public health issue in many developing countries, causing waterborne outbreaks as well as sporadic hepatitis. We report here an outbreak of HEV genotype 1f infection during Apr-May 2018 among persons living at Halisohor, a low land of southern part of Chottogram District of Bangladesh. A total of 933 patients were admitted in Combined Military Hospital (CMH) Chottogram with symptoms of acute hepatitis. Among them 550 patients were tested by ELISA for HEV specific IgM and all were positive. Genotyping, sequencing and phylogenetic analysis based on ORF 2 region revealed that the outbreak was caused by genotype 1f and the strains were closely related to the previously reported HEV strains that caused outbreak in Bangladesh in 2010. The current outbreak was most likely linked with water supply as fecal contamination in water was evident and could be prevented by ensuring access to safe drinking water.

Keywords: Hepatitis E virus, HEV, Bangladesh, Outbreak, genotype 1f

Hepatitis E virus (HEV) is a major cause of acute hepatitis disease worldwide, particularly in South Asia, including Bangladesh[1]. A population-based study conducted in rural communities in southern Bangladesh during 2003-05 reported that 22.5% of people had been previously infected with HEV and the incidence rate was 60 per 1000 persons-years[2]. Another study among the same population indicated that 19-27% of all maternal deaths and 7-13% of all neonatal deaths were associated with jaundice during pregnancy and a major proportion of these episodes were likely due to HEV[3].

HEV belongs to genera *Orthohepevirus A* under the family Hepeviridae. The virus has single serotype but is classified in eight genotypes: HEV1 and HEV2 cause large outbreaks in humans; HEV3 and HEV4 tend to be zoonotic and cause sporadic cases in humans; and HEV5-8 are confined in animals only[4]. Each genotype is subdivided into several subtypes; genotype 1 has six subtypes (a-f) for example. The most common circulating human HEV strains in South Asia including Bangladesh are subtypes 1a, 1c and 1f [5].

HEV is primarily transmitted through fecal-oral route, and outbreaks in endemic areas are typically associated with contaminated drinking water sources although less frequent person-to-person and vertical transmissions have also been reported[6]. The first laboratory confirmed HEV outbreak using retrospective samples was reported in Delhi during 1955-56. Since then, many HEV outbreaks were reported worldwide especially in Asia and Africa[6]. Due to absence of routine surveillance, only a few HEV outbreaks have been reported from Bangladesh and underrepresenting the importance of the disease[7, 8].

Currently, no antiviral drugs or HEV vaccines are being used in HEV-endemic areas. Only one hepatitis E vaccine, Hecolin-239 was licensed in China in 2012 but is not being used outside China[9]. A large phase IV trial of this vaccine is being conducted by icddr,b among more than 20,000 Bangladeshi women at their child bearing age (16 -39 years).

We report here an outbreak of HEV genotype 1f among persons living at Halisohor, a low land of southern part of Chottogram District of Bangladesh near to Bay of Bengal. This outbreak study was approved by the authority of the Combined Military Hospital (CMH) Chottogram, Govt. of Bangladesh.

Between 15 April and 30 May 2018, a total of 933 patients with acute jaundice were admitted to a 500 bedded Government hospital near Halisohor. Among them 911 (96.6 %) were adults (more than 18 years). The common symptoms were yellow coloration of urine, skin & eye, decreased appetite, vomiting and generalized weakness. High bilirubin (>3 mg/dl) was identified in 820 (88%); and elevated ALT (>125 mg/dl) in 790 (85%); ALP (>250 mg/dl) in 303 (32%) and PT (> 4 sec of normal) in 43 (5%) patients.

A total 550 patients were tested for HEV IgM using EIAgen HEV IgM Kit (Adaltis S.r.l, Rome, Italy) and all were found positive. All cases ultimately recovered well but required hospitalization: 387 (41.5%) patients for up to 2 weeks; 358 (38.4%) for 3 weeks; 130 (13.9%) for 4 weeks, and 58 (6.2%) for >4 weeks.

RNA was extracted from 34 randomly selected serum samples using QIAamp Viral RNA Minikit (Qiagen, Hilden, Germany) and screened for the presence of HEV RNA by real time PCR (Altona

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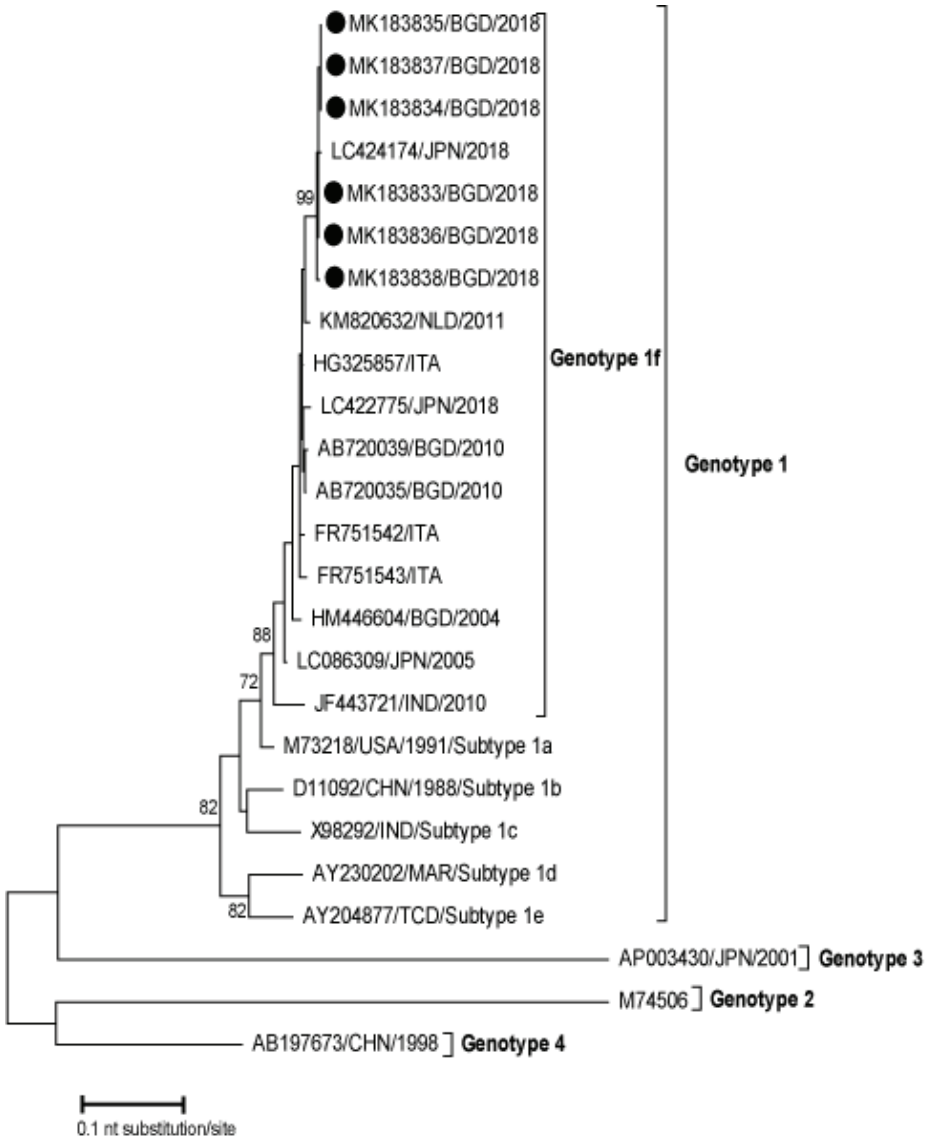

Figure 1. *Phylogenetic tree includes the partial ORF2 sequences of the current outbreak strains (indicated by closed circles) and previously identified global HEV genotype 1 strains. Reference genotypes (2-4) are included as outgroup. The evolutionary analysis was inferred by using the Maximum Likelihood method based on the Tamura-Nei model, using MEGA 7. A discrete Gamma distribution was used to model evolutionary rate differences among sites (5 categories (+G, parameter = 0.4507)). The numbers adjacent to the nodes represent the percentage of bootstrap support (of 1000 replicates) for the clusters to the right of the node. Bootstrap values lower than 70% are not shown.*

RealStar HEV RT- PCR kit, Germany). HEV RNA was positive in 27 (79%) samples. For genotyping, nucleotide sequencing was conducted for partial capsid gene (420 nucleotide bases) in the ORF2 [10] and submitteed to GenBank under accession number MK183833, MK183834, MK183835, MK183836, MK183837 and MK183838. All sequences were 99.8% identical to each other and were classified as genotype 1f using HEV genotyping tools (https://www.rivm.nl/mpf/typingtool/hev/).

The highest nucleotide identity using BLAST searches was found with strain 330106 (accession LC424174) which caused outbreak in Japan during 2016-17 and strains E13-Ban10 (accession AB720035) that caused outbreak in Bangladesh in 2010[8]. Phylogenetic analysis (Figure 1) revealed that the outbreak strains clustered very closely together in a monophyletic branch with Bangladeshi strains as well as recently identified strains from Japan, India, Italy and the Netherlands.

The outbreak was occurred in a defined area of about 4,000 habitats who use common municipal water for drinking and other purposes. Fecal contamination was confirmed in routine test by detecting thermotolerant fecal coliforms in water samples collected from different sources/reservoirs/tap points in that area during the outbreak. Further laboratory investigation to confirm HEV virus in water sources of the affected area was not possible since all the water sources were vigorously treated with chlorine just after the outbreak.

In summary, the recent jaundice outbreak in Bangladesh was caused by HEV genotype 1f which is genetically related to the previously identified indigenous HEV strain circulating in Bangladesh as well as global outbreak strains. The outbreak most likely linked with water supply and could be prevented by ensuring access to safe drinking water.

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References

1. Purcell RH, Emerson SU. Prevention. 2005. In: Thomas HC, Lemon S. Zuckerman AJ eds. *Viral hepatitis*, 3rd edn. Malden: Blackwell Publishing. 635-45.
2. Labrique AB, Zaman K, Hossain Z, Saha P, Yunus M, Hossain A, et al. 2010. Epidemiology and risk factors of incident hepatitis E virus infections in rural Bangladesh. *Am J Epidemiol.* **172** (8):952-61.
3. Gurley ES, Halder AK, Streatfield PK, Sazzad HM, Nurul Huda TM, Hossain MJ, et al. 2012. Estimating the burden of maternal and neonatal deaths associated with jaundice in Bangladesh: possible role of hepatitis E infection. *Am J Public Health.* **102**(12): 2248-54.
4. Sridhar S, Lo SK, Xing F, Yang J, Ye H, Chan JF, et al. 2017. Clinical characteristics and molecular epidemiology of hepatitis E in Shenzhen, China: a shift toward foodborne transmission of hepatitis E virus infection. *Emerg Microbes Infect*. **6**(12):e115.
5. Pathak R, Barde PV. 2017. Detection of Genotype 1a and 1f of Hepatitis E Virus in Patients Treated at Tertiary Care Hospitals in Central India. *Intervirology*. **60**(5): 201-6.
6. Hakim MS, Wang W, Bramer WM, Geng J, Huang F, de Man RA, Peppelenbosch MP, Pan Q. 2017. The global burden of hepatitis E outbreaks: a systematic review. *Liver Int*. Jan. **37**(1): 19-31. Epub 2016 Sep 9.
7. Gurley ES, Hossain MJ, Paul RC, et al. 2014. Outbreak of hepatitis E in urban Bangladesh resulting in maternal and perinatal mortality. *Clin Infect Dis*. **59**: 658-65.
8. Harun Or Rashid M, Akbar SMF, Takahashi K, Al Mahtab M, Khan MSI, Alim MA, et al. 2013. Epidemiological and molecular analyses of a non seasonal outbreak of acute icteric hepatitis E in Bangladesh. *J Med Virol*. **85** (8): 1369-76.
9. Zhu FC, Zhang J, Zhang XF, Zhou C, Wang ZZ, Huang SJ, et al. 2010. Efficacy and safety of a recombinant hepatitis E vaccine in healthy adults: a large-scale, randomised, double-blind placebo-controlled, phase 3 trial. *The Lancet*. **376** (9744): 895-902.
10. La Rosa G, Fratini M, Muscillo M, Iaconelli M, Taffon S, Equestre M, et al. 2014. Molecular characterisation of human hepatitis E virus from Italy: comparative analysis of five reverse transcription-PCR assays. *Virol J*. **11**(1): 72.