



## No molecular evidence of Hepatitis E infection among patients with HIV in Iran

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### Brief Communication

Hepatitis E virus (HEV) typically causes an acute self-limited infection in immunocompetent individuals. However, in recent years, multiple studies have demonstrated that immunocompromised patients such as transplant recipients, hematological patients, and HIV-infected patients are prone to acquiring chronic HEV infection and cirrhosis. In fact, chronic HEV infection in these high-risk groups can be considered as an emerging and clinically significant problem (1, 2). In general, HEV RNA-positive serum or stool for 6 months or longer has been suggested as a definition for chronic hepatitis E (3).

According to some previous studies, HIV-infected patients are at higher risk of acquiring chronic HEV infection than individuals without HIV (4). Immunosuppression can result in chronic HEV infection. So, infection with HIV is considered as one of the probable causes of HEV persistence (5). There are some evidences of acute HEV infections and persistent carriage of HEV in patients with HIV (6). Nevertheless, studies on HIV/HEV coinfection in HIV-infected patients are rare in Iran. The aim of present study was to determine the molecular prevalence of HEV infection in Iranian HIV-infected patients.

In total, 86 HIV-1 infected patients referred to hospitals of Tehran city (affiliated to Iran University of Medical Sciences, Tehran, Iran) participated in the current study. This work was performed between February 2015 and April 2016. Patients who had received anti-retroviral ther-

apy were excluded from the study. The project was approved by the ethics committee of Iran University of Medical Sciences, Tehran, Iran. The patients were informed about the aims of the study, and they signed informed consent before their enrollment. For sampling, approximately five ml of whole blood was taken from each candidate in an anti-coagulating manner. The plasma was isolated from the samples using centrifuge, and stored at -70 °C until use.

HEV-RNA was extracted from plasma samples using High Pure viral Nucleic Acid Kit (Roche Diagnostics, Germany) according to the manufacturer's protocol. cDNA synthesis was carried out using the first strand cDNA synthesis kit by Revert AidcDNA synthesis kit (Thermo Scientific, USA) according to the manufacturer's instruction. Detection of HEV-RNA performed by a TaqMan real-time RT-PCR assay (7). The primers (forward: 5'-GGTGGTTTCTGGGGTGAC-3' and reverse: 5' AGGGGTTGGTTGGATGAA-3') amplify a 70bp fragment in ORF2 of HEV genome. The amplified region is detectable using TaqMan probe (5'-FAM-TGATTCTCAG CCCTTCGC-BHQ-3'). HIV-1 RNA quantification was performed with the COBAS TaqMan (Roche Diagnostics, USA), according to the manufacturer's instructions.

The demographic data of the patients showed that 53 (61.6%) of the cases were males and 33 (38.4%) were females. The mean age was 38.23±9.58 years, with a wide

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#### ↑What is "already known" in this topic:

Previous studies have suggested that patients with HIV infection are at risk for acquiring HEV infection. Based on the several studies that have been conducted thus far, detection rate of HEV-RNA was low and ranges from 0 to 1.3% among HIV positive patients.

#### →What this article adds:

Our study has shown that none of the 86 HIV-positive patients tested had HEV RNA in their plasma examined by real-time RT-PCR assay, which was consistent with results of other previous studies.

range (4-65 years) of age distribution. The mean HIV viral load of all patients with HIV was 32026.75 copies per ml. None of the 86 HIV-positive patients tested had HEV-RNA detected in their plasma by real-time RT-PCR assay.

In the recent years, HEV infection has introduced as an emerging infection in immunosuppressed patients worldwide, most notably in resource-limited countries. It has been shown that HEV infection can lead to acute and chronic hepatitis in immunocompromised patients, including in patients with late stage of HIV infection. The presence of HEV-RNA in patient's specimen indicates an ongoing infection (8).

Based on the several studies that have been conducted thus far, the prevalence of IgG against hepatitis E virus among HIV positive cases ranges from 1.5-11.2% worldwide, while the detection rate of HEV-RNA is low and ranges from 0 to 1.3% (9). Our study has shown the lack of detectable HEV-RNA in plasma samples obtained from a group of Iranian HIV patients, which was consistent with results of other previous studies (10-17).

Only limited studies carried out on the prevalence of HEV infection among Iranian HIV-infected patients. Joulaei et al. reported that the overall seroprevalence of hepatitis E among patients infected with HIV was 16.4% (18). Another study by Ramezani et al. showed that the seroprevalence of HEV was 10% in HIV patients and no HIV-infected patients had detectable hepatitis E virus-RNA (15).

Detection of specific anti-HEV antibodies can help to determine whether the infection was recent; however, these antibodies can persist for months or even years after acute infection, without detection of any HEV-RNA. For this reason, the rate of HEV seroprevalence is usually significantly higher than HEV-RNA prevalence. On the other hand, the results of serological assays may be negative despite ongoing or occult HEV infection (19); hence, in patients with HIV infection, PCR assay for HEV-RNA should be considered.

In recent years, molecular assays such as real-time PCR analysis have been applied for viral genomic RNA detection and diagnosis of HEV infection (20, 21). It has demonstrated that the presence of HEV-RNA in fecal or serum specimens is the most reliable marker for diagnosis of HEV infection (22-24). Real-time PCR provides a rapid, sensitive, and robust method for measuring HEV-RNA that has replaced the traditional serological assays as the gold standard for diagnosis of HEV infection (25); however, duration of HEV viremia in primary infections is short (10-30 days after the onset of symptoms). So, detection of HEV RNA is difficult for diagnosis in clinical practice within the proper time. Nevertheless, it should be noted that our small sample size can be considered as a limitation of the study. On the other hand, some previous studies have reported an increased seroprevalence of HEV in immunocompromised patients with HIV infection. Therefore, further case-control studies with serology approach and larger sample size, alongside detection of HEV RNA in both blood and stool samples are recommended for future research.

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## Conflict of Interests

The authors declare that they have no competing interests.

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