

# Seroprevalence of the hepatitis E virus among blood donors in the Qassim Region, Saudi Arabia

Bader Alhatlani (✉ [balhatlani@qu.edu.sa](mailto:balhatlani@qu.edu.sa))

Qassim University <https://orcid.org/0000-0003-0871-6313>

Waleed Aljabr

King Fahad Medical City

Mohammed Almarzouqi

Qassim University Community College of Unaizah

Sami Alhatlani

King Saud Hospital

Rayan Alzunaydi

King Saud Hospital

Abeer Alsaykhan

King Saud Hospital

Sulaiman Almaiman

King Saud Hospital

Ahmed Aleid

King Fahad Medical City

Ammar Alsughayir

King Fahad Medical City

Yara Beshawri

King Fahad Medical City

Abdulrahman Almusallam

Qassim University Community College of Unaizah

---

## Research note

**Keywords:** hepatitis E virus, transfusion, ELISA, seroprevalence, serology

**Posted Date:** October 14th, 2020

**DOI:** <https://doi.org/10.21203/rs.3.rs-18073/v2>

**License:**   This work is licensed under a Creative Commons Attribution 4.0 International License.

[Read Full License](#)

# Abstract

## Objectives

To evaluate the seroprevalence of hepatitis E virus (HEV), a major public health issue worldwide with the potential for transmission via blood transfusion, in blood donors in the Qassim Region, Saudi Arabia. Serum samples (n = 1,078) were collected from volunteer blood donors from January to April 2019 and tested for the presence of anti-HEV IgG and IgM by indirect enzyme-linked immunosorbent assays.

## Results

Overall, the seroprevalence of anti-HEV IgG among blood donors was 5.7%. Additionally, the seropositive rates of anti-HEV IgG and IgM were significantly higher in non-Saudi donors (22.1% and 35.3%) than in Saudi donors (3% and 7.4%). The seroprevalence of anti-HEV IgG increased with age; however, there was no correlation between gender and anti-HEV IgG and/or IgM. The seroprevalence of HEV among blood donors in the Qassim Region was lower than previous estimates for other regions of the country. Further studies covering a wider geographical area are needed to validate and expand the findings and to determine the importance of HEV screening in the region.

## Introduction

Hepatitis E virus (HEV), belonging to the *Orthohepevirus* genus in the family *Hepeviridae*, causes liver diseases in humans [1]. The virus was first described in the early 1980s as a non-A and non-B hepatitis virus and was subsequently cloned in 1991 [2,3]. HEV contains a small nonenveloped, positive-sense single-stranded RNA genome approximately 7,200 nucleotides long [4]. Similar to hepatitis A, the vast majority of HEV infections are asymptomatic (especially in children) or cause self-limiting acute liver inflammation, which can resolve within a few weeks without any need for specific treatment [4]. However, immunosuppressed individuals, organ-transplant recipients, and pregnant women are at an increased risk of developing life-threatening diseases, including chronic hepatitis and acute liver failure, after HEV infection [5,6]. It has been estimated that the mortality rates for HEV infections in pregnant women and young people are about 20% and 3%, respectively [7]. The incubation period, which occurs during the prodromal phase, can vary from 2 to 8 weeks, and common symptoms of HEV infection during this period are usually nonspecific and include fever, nausea, vomiting, and malaise [8]. HEV is now recognized as a major public health issue, causing over 20 million infections every year worldwide and accounting for approximately 70,000 deaths [1]. Currently, there are eight known HEV genotypes from only one serotype that can infect humans and other animal taxa (HEV-1 to HEV-8), of which HEV-1 and HEV-2 are restricted to humans and are associated with most outbreaks in developing countries in parts of Asia and Africa [9]. Genotypes 3 and 4 are typically identified in developed countries, including the USA, New Zealand, Japan, and most countries in Europe, and can be isolated from a broader range of taxa, including humans, pigs, deer, and rabbits. Genotypes 5 and 6 are found in wild boars [8,10]. More

recently, a new genotype of camelid HEV (HEV-7) was isolated from dromedary camels in the UAE and some African countries, including Sudan and Egypt [11,12].

Fecal-oral transmission is considered the main route of HEV transmission, but other transmission routes have been suggested. This includes person-to-person transmission, such as vertical transmission (mother-to-infant) during delivery. In particular, blood transfusion transmission has become one of the main routes, especially in some low-income countries in Asia [4]. Several studies have reported transfusion-transmitted HEV from blood components in many industrialized countries, including European countries, Australia, and the United States [13-18]. In the Gulf and other neighboring countries, few studies have evaluated the HEV seroprevalence in healthy blood donors. A recent study has indicated that the HEV seroprevalence in Qatar is high among blood donors, i.e., approximately 21% [19]. To the best of our knowledge, the first study of HEV in Saudi Arabia was conducted in 1994 in Riyadh and Gizan, with anti-HEV antibody detection frequencies of 8.4% and 14.9%, respectively [20]. Two additional studies of HEV in Saudi Arabia reported seroprevalences in blood donation samples of 18.7% and 16.9% in Makkah and Jeddah, respectively [21,22]. In most countries, screening for HEV in blood donors is not conducted. However, screening was introduced in several European countries, and in Japan [4]. In addition, the United States is now considering HEV screening for blood donors [4]. In this study, we estimated the HEV seroprevalence among blood donors in the Qassim Region, Saudi Arabia. These results provide a basis for evaluating whether routine screening is necessary in Saudi Arabia.

## **Materials And Methods**

### **Study design and sample collection**

From January to April 2019, 1,078 whole blood samples were collected from volunteer blood donors at the Blood Donor Unit at King Saud Hospital, Unayzah, Qassim province, Saudi Arabia.

### **Serological testing**

Serum samples were tested for the presence of anti-HEV IgG and IgM antibodies using commercial HEV Enzyme-linked Immunosorbent Assay (ELISA) Kits (Fortress Diagnostics, Antrim, UK) according to the manufacturer's instructions. The sensitivity and specificity of the assays are 99.5% and 99.6%, respectively [23]. Samples were tested in duplicate and samples yielding borderline results were retested in duplicate to confirm the initial results. Only IgG-positive samples were tested for the presence of anti-HEV IgM.

### **Statistical analyses**

The Chi-square test and Fisher's exact test with the Freeman-Halton extension were used to evaluate the associations between the demographic characteristics of the participants and HEV seroprevalence. All statistical analyses were performed using an online statistical calculator at: (<https://www.socscistatistics.com>); a two-tailed *p*-value of 0.05 was considered significant.

# Results

Whole blood samples (n = 1,078) were collected from blood donors at King Saud Hospital in the Qassim Region, Saudi Arabia. General characteristics of the blood donors are summarized in Table 1. The study population included 1,002 (93%) men and 76 (7%) women, with 924 (85.7%) Saudis and 154 (14.3%) non-Saudis with different nationalities. The age of participants ranged from 18 to 73 years (mean  $\pm$  SD, 34.5  $\pm$  10.3 years); 461 donors (42.8%) were aged 25 to 34 years, 202 donors (18.7%) were younger than 25 years, and 61 donors (5.7%) were older than 55 years (Table 1).

**Table 1. Characteristics of the study population and anti-HEV IgG results.**

<u>Gender</u>	<u>Total (n = 1,078)</u>	<u>HEV IgG-positive (n = 61)</u>	<u>HEV IgG-negative (n = 1,017)</u>	<u>p-value</u>
Female	76 (7%)	2 (2.6%)	74 (97.4%)	0.24
Male	1,002 (93%)	59 (5.9%)	943 (94.1%)	
<u>Nationality</u>				
Saudi	924 (85.7%)	27 (3%)	897 (97%)	<0.001
Non-Saudi	154 (14.3%)	34 (22.1%)	120 (77.9%)	
<u>Age group (years)</u>				
<25				0.01
25-34	202 (18.7%)	2 (1%)	200 (99%)	
35-44	461 (42.8%)	28 (6%)	433 (94%)	
45-54	247 (22.9%)	16 (6.5%)	231 (93.5%)	
$\geq$ 55	107 (9.9%)	11 (10.3%)	96 (89.7%)	
	61 (5.7%)	4 (6.5%)	57 (93.5%)	

In total, 61 of the 1,078 blood samples (5.7%) were positive for anti-HEV IgG, including 2 samples from women (2.6%) and 59 from men (5.9%), with no significant difference between males and females ( $p = 0.24$ ). However, the anti-HEV IgG seroprevalence was significantly higher in non-Saudis (22.1%) than in Saudis (3%) (Table 1,  $p < 0.001$ ). We also analyzed the positive anti-HEV IgG blood donor samples for the seroprevalence of anti-HEV IgM. Our results indicated that 14 of the 61 anti-HEV IgG positive samples (23%) were positive for anti-HEV IgM (Table 2). Again, the seroprevalence of anti-HEV IgM was significantly higher in blood samples from non-Saudi donors (35.3%) than in those from Saudi donors (7.4%) (Table 2,  $p < 0.001$ ). There were no significant differences were found in the frequencies of anti-HEV IgM-positive samples between women and men ( $p = 0.99$ ).

**Table 2. Characteristics of the study population and anti-HEV IgM results.**

<u>Gender</u>	<u>Total</u>	<u>HEV IgM-positive</u>	<u>HEV IgM-negative (n = 47)</u>	<u>p-value</u>
-	<u>(n = 61)</u>	<u>(n = 14)</u>	1 (50%)	-
Female	2 (3.3%)	1 (50%)	47 (78%)	0.99
Male	59 (96.7%)	13 (22%)		
<u>Nationality</u>				
Saudi	27 (44.3%)	2 (7.4%)	25 (92.6%)	<0.001
Non-Saudi	34 (55.7%)	12 (35.3%)	22 (64.7%)	
<u>Age group (years)</u>				
<25				
25-34	4 (6.6%)	2 (50%)	2 (50%)	0.72
35-44	26 (42.6%)	6 (23%)	20 (77%)	
45-54	18 (29.5%)	5 (27.8%)	13 (97.2%)	
≥55	7 (11.5%)	1 (14.3%)	6 (85.7%)	
	6 (9.8%)	0 (0%)	6 (100%)	

Furthermore, we found a significantly higher rate of HEV IgG-positive samples for donors between 45 to 54 years old (10.3%,  $p = 0.01$ ) than for other age groups, while donors younger than 25 years old had only an anti-HEV IgG seropositive rate of 1% (Fig 1). The other age groups had similar anti-HEV IgG seropositive rates, and there was no significant association between age groups and anti-HEV IgM seropositivity ( $p = 0.72$ ).

Of the 14 anti-HEV IgM-positive serum samples, only 2 samples were positive for anti-HCV antibodies, both of which were from male non-Saudi donors. There was no significant association between anti-HEV and antibodies against hepatitis C virus (anti-HCV). In addition, none of the samples was positive for antibodies against the surface antigens of the hepatitis B virus (anti-HBsAg), antibodies against human immunodeficiency virus (anti-HIV), and antibodies against human T-lymphotropic virus type I and II (anti-HTLV I/II).

## Discussion

HEV is a transfusion-transmissible virus, as evidenced by its detection in blood donors in both developed and developing countries [24,25]. In the current study, we screened 1,078 serum samples from blood donors in the Qassim Region, Saudi Arabia for anti-HEV IgG and IgM antibodies. This is the largest study of the seroprevalence of HEV in Saudi Arabia to date with respect to the number of samples obtained from blood donors. Our results indicated that the seroprevalence of HEV in blood donors is 5.7% for IgG. In addition, we detected a significant difference between Saudis (3%) and non-Saudis (22.1%) in terms of seroprevalence of anti-HEV IgG. The seroprevalence of anti-HEV IgM was also significantly higher among non-Saudi blood donors (35.3%) than in Saudi blood donors (7.4%). The age of non-Saudi blood donors who tested positive for anti-HEV IgG ranged from 26 to 62 years (mean  $\pm$  SD, 36.9  $\pm$  6.3 years), whereas the age of Saudi blood donors who tested positive for anti-HEV IgG ranged from 21 to 61 years (mean  $\pm$  SD, 39.1  $\pm$  9.7 years). We also found an increase in the rate of anti-HEV IgG with age. The highest rate of positive anti-HEV IgG results was observed in the 45- to 54-year-old group and the lowest rate was obtained for donors younger than 25 years (Fig 1). These results are consistent with those of a previous study suggesting that age is a risk factor for HEV [26]. The anti-HEV IgM seroprevalence was comparable

in all age groups other than donors aged over 55 years who tested negative for IgM. Some studies have suggested that the HEV seroprevalence rate increases with age, while other studies have reported that the correlation between age groups and HEV seroprevalence varies [13, 22]. The anti-HEV IgG seroprevalence among blood donors in the Qassim Region was comparable to that in Riyadh (8.4%) but was much lower than estimates for other regions of Saudi Arabia, such as Gizan (14.9%), Makkah (18.7%), and Jeddah (16.9%) [20-22]. Moreover, the seroprevalence of anti-HEV IgG was also lower than that reported in neighboring countries, such as Qatar (20.7%) [19]. This variation in the HEV seroprevalence among studies can be explained by differences in the commercial enzyme immunoassays used to detect HEV antibodies in blood donor samples. Commercial ELISA assays have different sensitivities and specificities, which may lead to variation in estimates of the HEV seroprevalence [23]. For instance, the HEV ELISA kit used in the present study (Fortress Diagnostics) has a sensitivity and specificity of 99.5% and 99.6%, respectively, but the most common ELISA for HEV detection (the Wantai assay) is known for its high antibody detection rates [24]. In addition, these assays may not detect all HEV genotypes. The variation among studies may also be explained by differences in demographic properties, which can vary among cities within the same country. For example, the higher HEV seroprevalence in Makkah and Jeddah compared to other cities in Saudi Arabia could be due to the high number of visitors of different nationalities in these two cities every year for pilgrimages and other religious practices.

In summary, we showed that the seroprevalence of HEV is relatively low among blood donors in the Qassim Region, Saudi Arabia, when compared with that in other regions of the country. The anti-HEV IgG seropositivity was significantly higher in non-Saudi donors (22.1%) than in Saudi donors (3%). Further investigations of the seroprevalence of HEV in a larger sample of blood donors from an expanded geographical distribution are needed to determine the need for HEV screening for blood donors in Saudi Arabia.

## Limitations

A potential limitation of the current study is that we could not confirm the presence of HEV RNA in the seropositive donor samples. Another limitation is that women are underrepresented in this study due to the fewer number of women blood donors compared with men donors at the Blood Donor Unit at King Saud Hospital, Unayzah, Qassim province, Saudi Arabia.

## Abbreviations

**Anti-HBsAg:** Antibodies against the surface antigens of the hepatitis B virus

**Anti-HCV:** Antibodies against hepatitis C virus

**Anti-HIV:** Antibodies against human immunodeficiency virus

**Anti-HTLV I/II:** Antibodies against human T-lymphotropic virus type I and II

**ELISA:** Enzyme-linked immunosorbent assay

**HEV:** Hepatitis E virus

## **Declarations**

### **Ethics approval and consent to participate**

All study participants provided written informed consent, and the study design was approved by the local research Ethical Committee of the General Directorate of Health Affairs at Qassim Province (approval number: 1441-225162).

### **Consent for publication**

Not applicable.

### **Availability of data and materials**

The datasets used and/or analysed during the current study are available from the corresponding authors on reasonable request.

### **Competing interests**

The authors declare that they have no competing interests.

### **Funding**

The authors gratefully acknowledge Qassim University, represented by the Deanship of Scientific Research, for the material support for this research under the number (3948-ucc-2018-1-14-S) during the academic year 2018-2019.

### **Authors' contributions**

BA and WA designed the study, analyzed and interpreted the results, and drafted the manuscript. MA, SA, RA, AA, ABAA and SHA collected the samples and performed experiments. AHAA, AHA, and YB conducted the statistical analysis. All authors read and approved the final manuscript.

### **Acknowledgments**

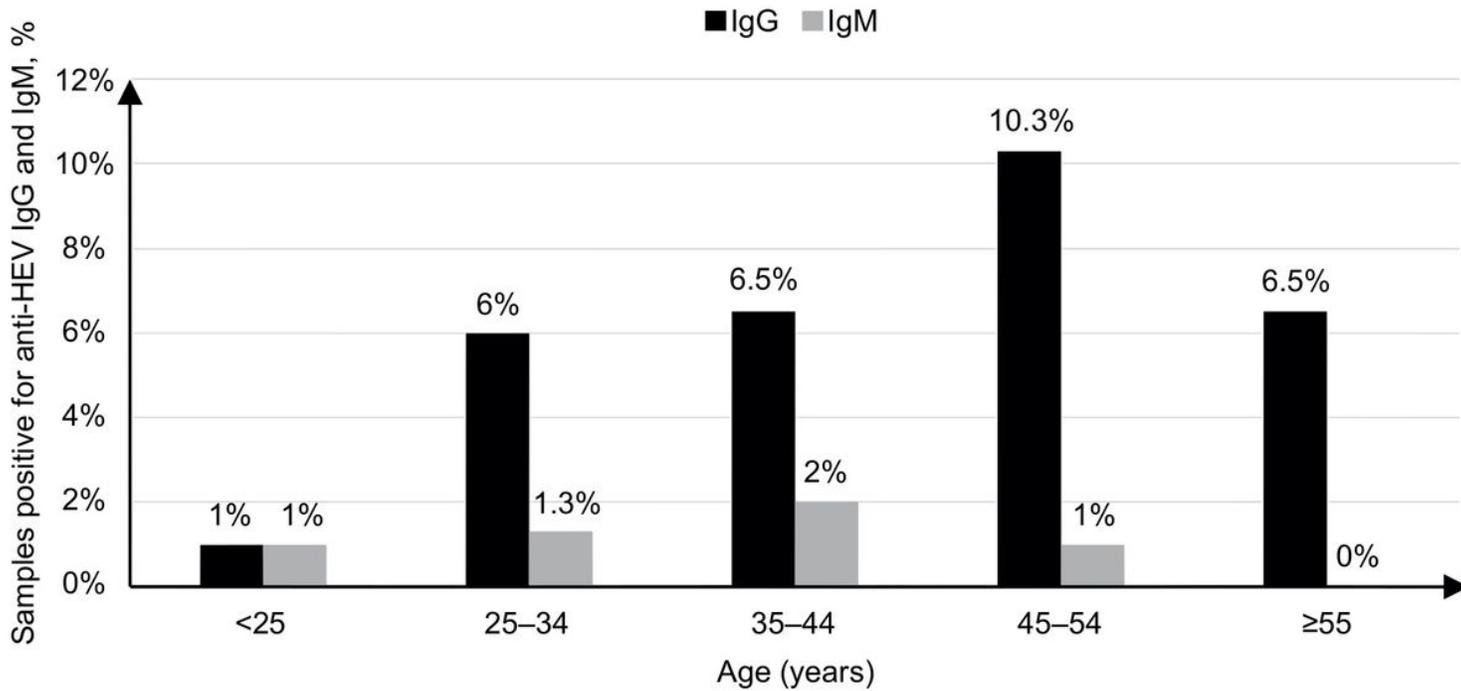
The authors thank all the staff in the Medical Laboratory Department at King Saud Hospital for their contribution to this study. We also would like to thank Editage ([www.editage.com](http://www.editage.com)) for English language editing.

## **References**

1. Debing Y, Moradpour D, Neyts J, Gouttenoire J. Update on hepatitis E virology: Implications for clinical practice. *J Hepatol*. 2016;65(1): 200–212. doi: 10.1016/j.jhep.2016.02.045.
2. Balayan MS, Andjaparidze AG, Savin Skaya SS, Ketiladze ES, Braginsky DM, Suvinov AP, et al. Evidence for a virus in non-A, non-B hepatitis transmitted via the fecal-oral route. *Intervirology*. 1983;20: 23–31. doi: 10.1159/000149370
3. Reyes GR, Purdy MA, Kim JP, Luk KC, Young LM, Fry KE, et al. Isolation of a cDNA from the virus responsible for enterically transmitted non-A, non-B hepatitis. *Science* 1990;247: 1335–1339. doi: 10.1126/science.2107574.
4. Kamar N, Izopet J, Pavio N, Aggarwal R, Labrique A, Wedemeyer H, et al. Hepatitis E virus infection. *Nat Rev Dis Primers*. 2017;3:17086. doi: 10.1038/nrdp.2017.86.
5. Kamar N, Selves J, Mansuy J-M, Ouezzani L, Péron J-M, Guitard J, et al. Hepatitis E virus and chronic hepatitis in organ-transplant recipients. *N Engl J Med*. 2008;358: 811–817. doi: 10.1056/NEJMoa0706992.
6. Kamar N, Dalton HR, Abravanel F, Izopet J. Hepatitis E virus infection. *Clin Microbiol Rev*. 2014;27: 116–138. doi: 10.1128/CMR.00057-13.
7. Mushahwar IK. Hepatitis E virus: Molecular virology, clinical features, diagnosis, transmission, epidemiology, and prevention. *J Med Virol*. 2008;80: 646–658. doi: 10.1002/jmv.21116.
8. Pérez-Gracia MT, García M, Suay B, Mateos-Lindemann ML. Current knowledge on hepatitis E. *J Clin Transl Hepatol*. 2015;3: 117–126.
9. Sooryanarain H, Meng XJ. Hepatitis E virus: reasons for emergence in humans. *Curr Opin Virol*. 2019;34: 10–17. doi: 10.1016/j.coviro.2018.11.006.
10. Forni D, Cagliani R, Clerici M, Sironi M. Origin and dispersal of Hepatitis E virus. *Emerg Microbes Infect*. 2018;7(1): 11. doi: 10.1038/s41426-017-0009-6.
11. Woo PCY, Lau SKP, Teng JLL, Tsang AKL, Joseph M, Wong EYM, et al. New hepatitis E virus genotype in camels, the Middle East. *Emerg Infect Dis*. 2014;20: 1044–1048. doi: 10.3201/eid2006.140140.
12. Sridhar S, Teng JLL, Chiu TH, Lau SKP, Woo PCY. Hepatitis E virus genotypes and evolution: Emergence of camel hepatitis E variants. *Int J Mol Sci*. 2017;18: E869. doi: 10.3390/ijms18040869.
13. Kaufmann A, Kenfak-Foguena A, André C, Canellini G, Bürgisser P, Moradpour D, et al. Hepatitis E virus seroprevalence among blood donors in Southwest Switzerland. *PLoS One*. 2011;6: e21150. doi: 10.1371/journal.pone.0021150
14. Vollmer T, Diekmann J, Johne R, Eberhardt M, Knabbe C, Dreier J. Novel approach for detection of hepatitis E virus infection in German blood donors. *J Clin Microbiol*. 2012;50: 2708–2713. doi: 10.1128/JCM.01119-12.
15. Gallian P, Lhomme S, Piquet Y, Sauné K, Abravanel F, Assal A, et al. Hepatitis E virus infections in blood donors, France. *Emerg Infect Dis*. 2014;20: 1914–1917.  
<https://doi.org/10.3201/eid2011.140516>.

16. Pawlotsky JM. Hepatitis E screening for blood donations: an urgent need? *Lancet*. 2014;384: 1729–1730. doi: 10.1016/S0140-6736(14)61187-9.
17. Shrestha AC, Shrestha AC, Seed CR, Flower RLP, Rooks KM, Keller AJ, et al. Hepatitis E virus and implications for blood supply safety, Australia. *Emerg Infect Dis*. 2014;20: 1940–1942. doi: 10.3201/eid2011.140412.
18. Ticehurst JR, Pisanic N, Forman MS, Ordak C, Heaney CD, Ong E, et al. Probable transmission of hepatitis E virus (HEV) via transfusion in the United States. *Transfusion*. 2019;59: 1024–1034. doi: 10.1111/trf.15140.
19. Nasrallah GK, Al Absi ES, Ghandour R, Ali NH, Taleb S, Hedaya L, et al. Seroprevalence of hepatitis E virus among blood donors in Qatar (2013-2016). *Transfusion*. 2017;57: 1801–1807. doi: 10.1111/trf.14116.
20. Arif M, Qattan I, al-Faleh F, Ramia S. Epidemiology of hepatitis E virus (HEV) infection in Saudi Arabia. *Ann Trop Med Parasitol*. 1994;88: 163–168. doi: 10.1080/00034983.1994.11812854.
21. Abdelaal M, Zawawi TH, al Sobhi E, Jeje O, Gilpin C, Kinsara A, et al. Epidemiology of hepatitis E virus in male blood donors in Jeddah, Saudi Arabia. *Ir J Med Sci*. 1998;167: 94–96. doi: 10.1007/bf02937946.
22. Johargy AK, Mahomed MF, Khan MM, Kabrah S. Anti-hepatitis E virus seropositivity in a group of male blood donors in Makkah, Saudi Arabia. *J Pak Med Assoc*. 2013;63: 185–189.
23. Al-Sadeq DW, Majdalawieh AF, Mesleh AG, Abdalla OM, Nasrallah GK. Laboratory challenges in the diagnosis of hepatitis E virus. *J Med Microbiol*. 2018;67: 466–480. doi: 10.1099/jmm.0.000706.
24. Nelson KE. Transfusion transmission of hepatitis E virus: an emerging issue. *Ann Blood*. 2017;2: 1-7. doi: 10.21037/aob.2017.09.02.
25. Capai L, Charrel R, Falchi A. Hepatitis E in high-income countries: What do we know? And what are the knowledge gaps? *Viruses*. 2018;10: 1–23. doi: 10.3390/v10060285.
26. Dalton HR, Bendall R, Ijaz S, Banks M. Hepatitis E: an emerging infection in developed countries. *Lancet Infect Dis*. 2008;8: 698–709. doi: 10.1016/S1473-3099(08)70255-X.

## Figures



**Figure 1**

Age distributions of 1,078 blood donors positive for anti-HEV IgG and/or IgM.