Estimating the global prevalence of hepatitis E virus in swine and pork products

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ABSTRACT

Zoonotic transmission of hepatitis E virus (HEV), in particular the genotype (GT) 3 and GT4 strains, constitutes a major one health issue. Swine serves as an important reservoir and the processed pork products essentially contribute to foodborne transmission. This study comprehensively estimated HEV prevalence in domestic pigs, wild boars, and pork products. At global level, we found nearly 60% domestic pigs and 27% wild boars have ever encountered HEV infection based seroprevalence rate. Nearly 13% domestic and 9.5% wild swine are actively infected based on HEV RNA positivity. Importantly, about 10% of commercial pork products are HEV RNA positive, although available data are limited in this respect. Our results indicate the high prevalence rate of HEV infection in pigs and widespread contamination in pork products, although there are substantial variations at regional and country levels. These findings are important for better understanding the global epidemiology and clinical burden of HEV infection in human population related to zoonotic transmission.

1. Introduction

Hepatitis E virus (HEV) is a non-enveloped, single-stranded positive-sense RNA virus. It is recognized as the leading cause of acute viral hepatitis. Globally, it is estimated approximately 939 million corresponding to 1 in 8 individuals have ever been infected with HEV [1]. Among the different HEV genotypes (GT) that affect human health, GT3 and GT4 are zoonotic, which have been found in various animal species [2]. Although HEV infection is usually self-limiting or asymptomatic in healthy individuals, GT3 and GT4 HEV infection in organ transplant patients is prone to develop chronic hepatitis [3,4].

Pigs serve as the major reservoir for the zoonotic HEV strains. Anti-HEV antibodies have been widely detected in both domestic pigs and wild boars [5–7]. There are different routes of HEV transmission from pigs to humans, such as direct contact with the animal, indirectly through contaminated environment and the consumption of pork products. However, the contribution of these different transmission routes can vary tremendously among different settings attributing to multi-factors, such as socioeconomic status, farming systems, food chains and life styles. Nevertheless, the widespread consumption of pork products is inevitably posing a major risk of HEV foodborne transmission in public health. Hepatitis E cases linking to consuming undercooked pork or wild boar meat have been widely reported [8,9].

Globally, the epidemiological feature and clinical burden of HEV infection in human population are distinct among different countries/regions [1]. It is intriguing to postulate whether this is associated with the specific prevalence rate of HEV in local swine population and available pork products. In this study, we aim to estimate the global prevalence of HEV in both domestic pigs and wild boars, as well as pork products in retailers.

2. Methods

2.1. Data sources, search strategies and study selection

A systematic search was conducted in Medline, Embase, Web of science, Cochrane CENTRAL and Google scholar. Databases were searched for articles in English language from inception until 31 May 2021. Studies were included if they contained epidemiological data about HEV in domestic pig or wild boar. The full search strategies and

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study selection criteria are provided in the Supplementary file S1-S2.

### 2.2. Data extraction, quality assessment and statistical analysis

Eligible studies were further divided into three study populations: domestic swine, wild boars and market/retailer pork products. Studies were scored according to Joanna Briggs Institute checklist for prevalence studies [10]. A 95% confidence interval (95% CI) was estimated using Wilson score method, and pooled prevalence rate was calculated by the DerSimonian-Laird random-effects model with Freeman-Tukey double arc sine transformation. Funnel plots and Egger regression test were used to assess potential publication bias. ‘M eta’ package in the R-3.5.3 statistical software was used for meta-analysis as previously described [11,12]. Sensitivity analysis was performed by
using ‘metainf’ to investigate the effects of group source and potentially unrepresentative samples. The details of quality assessment and statistical analysis are provided in supplementary S3.

3. Results and discussion

By comprehensively searching 5 databases (Supplementary S1–S3), we identified a total of 215 studies met the inclusion criteria, which were processed for analysis of HEV prevalence in domestic pigs, wild boars and pork products (sFig. 1). First, we estimated anti-HEV seroprevalence (indication of ever exposure) and HEV RNA positivity (indication of active infection) in domestic pigs. A total of 84 studies were included to estimate the global anti-HEV seroprevalence, resulting in a pooled rate of 59.33% (37 countries, 95% CI 53.64–64.90, $I^2 = 99\%$; sFig. 2). The highest seroprevalence was found in Oceania (75%, 95% CI 64.28–84.40), but this is only based on one study which likely causes bias in estimation. The second highest seroprevalence was found in Asia (67.45%, 95% CI 53.64–64.90, $I^2 = 99\%$; sFig. 2). The global estimation of HEV RNA positive rate was 12.71% (95% CI 10.81–14.73, $I^2 = 97\%$). The highest rate was found in North America (18.10%, 95% CI 8.71–29.84, $I^2 = 97\%$), followed by Europe (17.19%, 95% CI 13.16–21.61, $I^2 = 98\%$), South America (15.67%, 95% CI 6.75–27.33, $I^2 = 98\%$), Africa (12.29%, 95% CI 0.01–38.70, $I^2 = 99\%$), Asia (8.23%, 95% CI 6.21–10.49, $I^2 = 97\%$), and Oceania (6.52%, 95% CI 2.23–12.63) (sFigs. 4 and 5).

At country level, HEV prevalence in domestic pigs varies substantially, from 9.90% (Thailand, 95% CI 8.01–11.96) to 84.02% (India, 95% CI 44.05–100.00, $I^2 = 99\%$) of anti-HEV seroprevalence, and from 0% (Croatia, 95% CI 0.00–0.37) to 76.67% (Nigeria, 95% CI 67.32–84.89) of HEV RNA positivity (Table 1, Fig. 1). Importantly, we have collected genotyping information of swine HEV. GT3 is universally prevalent across the globe, whereas GT4 is mainly present in Western Pacific region. Interestingly, GT3 and GT4 are co-circulating in countries/territories, such as mainland China, Taiwan, Korea and Japan (Table 1, Fig. 1). This is consistent with clinical observations that both GT3 and GT4 HEV patients have been reported from these regions [13–16]. Although Europe is dominated by GT3, GT4 HEV has been identified in some peculiar cases including chronically infected patients [17]. Here, we found that GT3 and GT4 are also co-circulating in
increases over time, from 42.19% (95% CI 26.79–58.40) in domestic pig populations in Belgium (Fig. 1). Thus, the emergence of

Table 2

| Country | Anti-HEV antibodies | HEV RNA |
|---------|---------------------|--------|
|         | No. of studies | Events Tested (n) | Prevalence (%) | 95% CI | No. of studies | Events Tested (n) | Prevalence (%) | 95% CI |
| China   | 1             | 186 | 758 | 24.52 | 21.51–27.67 | 1           | 31 | 140 | 22.14 | 15.62 |
| Korea   | 1             | 1041 | 2736 | 38.05 | 36.24–39.88 | 1           | 24 | 1859 | 1.29 | 0.82–1.86 |
| Japan   | 6             | 262 | 1139 | 19.26 | 10.13–30.37 | 11         | 93 | 2609 | 3.20 | 2.51–3.95 |
| Thailand | 1           | – | – | – | – | 1           | 31 | – | 3.23 | 0.00–13.33 |
| Bulgaria | 1           | 98 | 240 | 40.83 | 34.68–47.13 | 1           | – | – | – | – |
| Croatia | 1             | 311 | 1000 | 31.10 | 28.27–34.01 | 1           | 17 | 150 | 11.33 | 6.70–16.95 |
| Czech   | 1             | 31 | 366 | 8.47 | 5.82–11.56 | 1           | – | – | – | – |
| Estonia | 1             | 81 | 471 | 17.20 | 13.92–20.75 | 1           | 13 | 81 | 16.05 | 8.77–24.93 |
| France  | 2             | 160 | 767 | 21.07 | 8.35–37.62 | 2           | 15 | 637 | 2.35 | 1.28–3.71 |
| Germany | 1             | 81 | 180 | 45.00 | 37.78–52.33 | 4           | 157 | 701 | 22.33 | 15.03–31.83 |
| Hungary | –             | – | – | – | – | 1           | 8 | 75 | 10.67 | 4.54–18.80 |
| Italy   | 6             | 560 | 3416 | 33.59 | 15.76–54.21 | 10          | 232 | 1697 | 13.56 | 6.57–22.46 |
| Lithuania | 1           | 178 | 312 | 57.05 | 51.51–62.50 | 1           | 86 | 505 | 17.03 | 13.87–20.44 |
| Netherlands | 1              | 293 | 1029 | 28.47 | 25.76–31.27 | 2           | 8 | 158 | 2.67 | 0.00–14.30 |
| Poland  | 1             | 90 | 290 | 31.03 | 25.83–36.49 | 1           | 12 | 120 | 20.00 | 12.38–27.67 |
| Portugal | –            | – | – | – | – | 1           | 24 | 120 | 20.00 | 12.38–27.67 |
| Romania | –             | – | – | – | – | 1           | 9 | 50 | 18.00 | 8.41–30.02 |
| Slovenia | 1           | 87 | 288 | 30.21 | 25.03–35.65 | 1           | 1 | 288 | 0.35 | 0.00–1.49 |
| Spain   | 4             | 409 | 1299 | 41.51 | 25.30–50.72 | 2           | 43 | 296 | 14.47 | 6.84–24.86 |
| Sweden  | –             | – | – | – | – | 1           | 13 | 159 | 8.18 | 4.36–13.03 |
| Switzerland | 1         | 38 | 303 | 12.54 | 9.03–16.52 | 1           | 3 | 137 | 0.00–1.84 | – |
| Turkey  | 1             | 0 | 93 | 0.00 | 0.00–1.84 | –           | – | – | – | – |
| EU/total | 1           | 121 | 104 | 11.54 | 6.02–18.47 | 1           | 1 | 104 | 3.85 | 0.83–8.58 |

Fig. 2. Subgroup analysis of anti-HEV seroprevalence or HEV RNA positivity.

domestic pig populations in Belgium (Fig. 1). Thus, the emergence of

Given the important role of foodborne transmission, we collected data on HEV RNA detection rates of pork meat, liver and sausage in retailers. This generated pooled positive rate of 9.5% (95% CI 21.69–32.28, I² = 98%) (Table 2, Fig. 8). Based on 37 studies from 18 countries/territories, the pooled rate of HEV RNA positivity was 9.45% (95% CI 6.42–12.96, I² = 96%) (Table 2, Fig. 9).

Considering the clear differences in husbandry and natural habitat between wild and domesticated pigs, we separately estimated HEV prevalence in wild boars. Based on data extracted from 33 studies from 19 countries/territories, we estimated that the overall anti-HEV seroprevalence was 26.82% (95% CI 21.69–32.28, I² = 98%) (Table 2, Fig. 8). Based on 37 studies from 18 countries/territories, the pooled rate of HEV RNA positivity was 9.45% (95% CI 6.42–12.96, I² = 96%) (Table 2, Fig. 9).
pork sausage. Notably, we estimated a nearly 3-fold HEV positivity rate of 15.23% (95% CI 11.62–19.21, $I^2 = 0\%$) in liver sausage, compared with 5.54% (95% CI 0.19–15.20, $I^2 = 82\%$) in pork sausage (Fig. 2, sFigs. 11–12).

Finally, we performed sensitivity analysis for HEV prevalence in domestic pigs and wild boars. In this meta-analysis, no significant change was observed by arbitrarily excluding any study from these groups. This low sensitivity supports the reliability of our estimation. However, funnel plot and Egger’s test indicate the presence of publication bias ($p > 0.05$) in three analyses, including seroprevalence and HEV RNA prevalence among domestic swine, and RNA prevalence among wild boars, which may potentially compromise the accuracy of prevalence estimation (sFigs. 13–26). Another limitation of our study is that we were unable to estimate HEV prevalence in pork products at regional/country levels and clarify the original place of the products, due to limited data available. Because the current food production and supply chains are diverse and complicated; it has become increasingly important to trace the origin of the contaminated products.

In summary, we found nearly 60% domestic pigs and 27% wild boars have ever encountered HEV infection at global level. Nearly 13% domestic and 9.5% wild swine are experiencing active infection. The risk of potential foodborne transmission is highlighted by our estimation that around 10% commercial pork products are HEV RNA positive. However, there remains gaps of translating these knowledge for better understanding the global epidemiology and clinical burden of HEV infection in human population related to zoonotic transmission. Because HEV zoonosis also involves many other factors, including socioeconomic status, farming style, food production and supply, as well as life styles. Nevertheless, our findings have set a stage for future research to further study the role of swine related HEV zoonosis and to facilitate the development of intervention and prevention strategies.

**Author contributions**

P. L., Z. M. and Q. P.: Project conceptualization. P. L., Y. L. and Y. J.: Data analysis. P. L. and Q. P.: Manuscript writing. Z. M. and Q. P.: Supervised the project. All authors reviewed and approved the manuscript.

**Ethical approval**

The authors confirm that the ethical policies of the journal, as noted on the journal’s author guidelines page, have been adhered to.

**Data availability statement**

All data needed to evaluate the conclusions in the paper are present in the paper and/or the Appendix.

**Declaration of Competing Interest**

The authors do not have any disclosures to report.

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**Appendix A. Supplementary data**

Supplementary data to this article can be found online at https://doi.org/10.1016/j.onehlt.2021.100362.

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