Zoonotic Aspects of Hepatitis E Virus in Denmark - DTU Orbit (23/12/2018)

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In this thesis the epidemiology of hepatitis E virus (HEV) in Denmark was explored. Globally, four genotypes of HEV are recognized along with several species specific variants. Non-zoonotic genotypes of the virus are found in the developing world, where they cause epidemics due to fecal contaminated water resources. Zoonotic genotypes have a primary reservoir in pigs worldwide. Humans infected with HEV are often asymptomatic, but can experience an acute self-limiting hepatitis. Pigs are asymptomatic and only mild lesions have been observed in the liver of infected animals. In the developed world, sporadic cases of HEV induced disease have been more frequently reported as awareness of the disease increases, including human cases in Denmark where HEV is also prevalent in pigs.

The thesis consists of an introduction into HEV where the literature on specific subjects is reviewed. The results obtained during the work of this PhD are presented in the form of four manuscripts. Finally, the four manuscripts are discussed in a broader context.

In Manuscript I, the work performed to investigate if HEV was found in other animal reservoirs than pigs in Denmark is presented. The research focused on mink (Nevison vison), an economically important livestock in Denmark. The main finding was the discovery of a novel variant of HEV. The virus was found in four different locations in samples collected in 2008 and in samples collected in 2011 indicating that the virus is widespread and has been circulating in minks for years. The virus did not seem to cause clinical disease in mink, however, this should be investigated further.

Manuscript II describes the teaching of HEV along with rotavirus (RV), porcine circovirus type 2 (PCV2), somatic coliphages, E. coli and Enterococcus spp. through field soil into tile drains under natural conditions of field fertilization. The microorganisms were naturally present in the pig slurry applied to the field. The results showed that HEV along with the other five microorganisms were found in water drained from the field, indicating potential contamination of water reservoirs in connection with the untreated drainage runoff. Rotavirus was detected in deeper groundwater screen indicating the possibility of groundwater contamination of viruses originating from manure, posing a risk for the contamination of important drinking water reservoirs in Denmark.

Manuscript III focuses on the viral contamination of mussels farmed in Denmark. The mussels was assayed for HEV and Rotavirus, known to occur in shellfish and cause disease. Furthermore, PCV2was also assayed as an indicator of porcine waste contamination. All samples analyzed were negative for HEV and RV, however, a large proportion of the samples tested positive for the PCV2. This is the first report that shows the potential of PCV2 as an indicator organism. The lack of HEV in the mussels is in accordance with previous studies, although shellfish has been reported as the source of multiple sporadic cases of HEV infection.

Manuscript IV report the results of a longitudinal study performed in a multi-site farm, where HEV development in 104 pigs were followed from farrowing to 17 weeks of age. The pigs were divided into three groups according to the level of antibody titers of their sows. During the study, successful transfers of maternal antibodies were observed only for the pigs born to sows with the highest antibody level. Furthermore, a significant reduction of the number of pigs shedding HEV from approx. 70% to 50% was observed in the group that received maternal antibodies. Ten of the pigs that were shedding HEV at week 17 were necropsied at week 20 and three of these were still shedding HEV. Correspondingly, HEV was found in different tissues, e.g. liver, tonsils and lungs, of these three pigs. HEV was, however, not found in muscles. In addition, high level of HEV was found in 1 out of 73 Danish livers purchased at grocery stores in the larger Copenhagen area.

Based on these studies it could be concluded that HEV belongs to a diverse family of viruses with variants in multiple species, and there is a possibility that many more of these will be identified during the coming years. However, clinical illness have so far only been described in humans and birds. HEV is known to be present in the water environment of poorly sanitized regions of the world, however, our study also emphasized the need for better understanding of viral leaching as well as the need for diagnostic tools enabling survey of water to maintain high drinking water quality also in the developed part of the world. We did not find HEV or RV in shellfish although indications of porcine waste contamination of shellfish occurring were observed in though the detection of the porcine specific virus PCV2. This is an interesting finding and suggest that PCV2 may be utilized to monitor viral contamination of shellfish, which is completely lacking today. Lastly, high levels of maternal antibodies can diminish the number of pigs shedding or carrying infectious HEV when entering the food supply chain. This could be utilized by vaccinating sows prior to farrowing, ensuring transfer of maternal antibodies and thereby reducing human exposure to HEV.

General information
State: Published
Organisations: National Veterinary Institute, Section for Virology, National Food Institute, Division of Food Microbiology
Contributors: Krog, J. S., Breum, S. Ø., Larsen, L. E., Schultz, A. C.
Number of pages: 159
Publication date: 2013

Publication information
Place of publication: Kgs. Lyngby
Publisher: Technical University of Denmark (DTU)
Original language: English
Electronic versions:
Ph.d._afhandling_Jesper_Schak_Krog_PDF
Research output: Research › Ph.D. thesis – Annual report year: 2013