

CROSSING BARRIERS: THE CASE OF RABBIT HAEMORRHAGIC DISEASE VIRUS-2

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BACKGROUND

Rabbit haemorrhagic disease

- Fatal, necrotizing hepatitis in *Oryctolagus cuniculus*
- Caused by pathogenic lagovirus: non-enveloped, single stranded positive sense RNA virus, Family *Caliciviridae*,)
- First reported in China in 1984, Europe in 1986
- Prior to 2010, only RHDV and RHDVa recognized



BACKGROUND

Rabbit Haemorrhagic Disease Virus 2 (RHDV2 or RHDVb):

- First reported in France in 2010
- Causes similar fatal, necrotizing hepatitis and hemorrhagic disease
- RHDV2 has rapidly crossed several notable barriers:
 - Geographic
 - Immunologic
 - Epidemiologic
- And created other barriers:
 - Analytical
 - Nomenclature



GEOGRAPHIC BARRIERS: TRANSMISSION

- Direct spread
- Indirect spread via infectious material carried by insects, predators, scavengers, humans
- Lagoviruses are highly infectious and persistent in environment
 - >7 months chilled in organic material
 - 3+ months dried on cloth
 - A single flyspot enough (2-3 LD₅₀ units)

GEOGRAPHIC SPREAD

2010



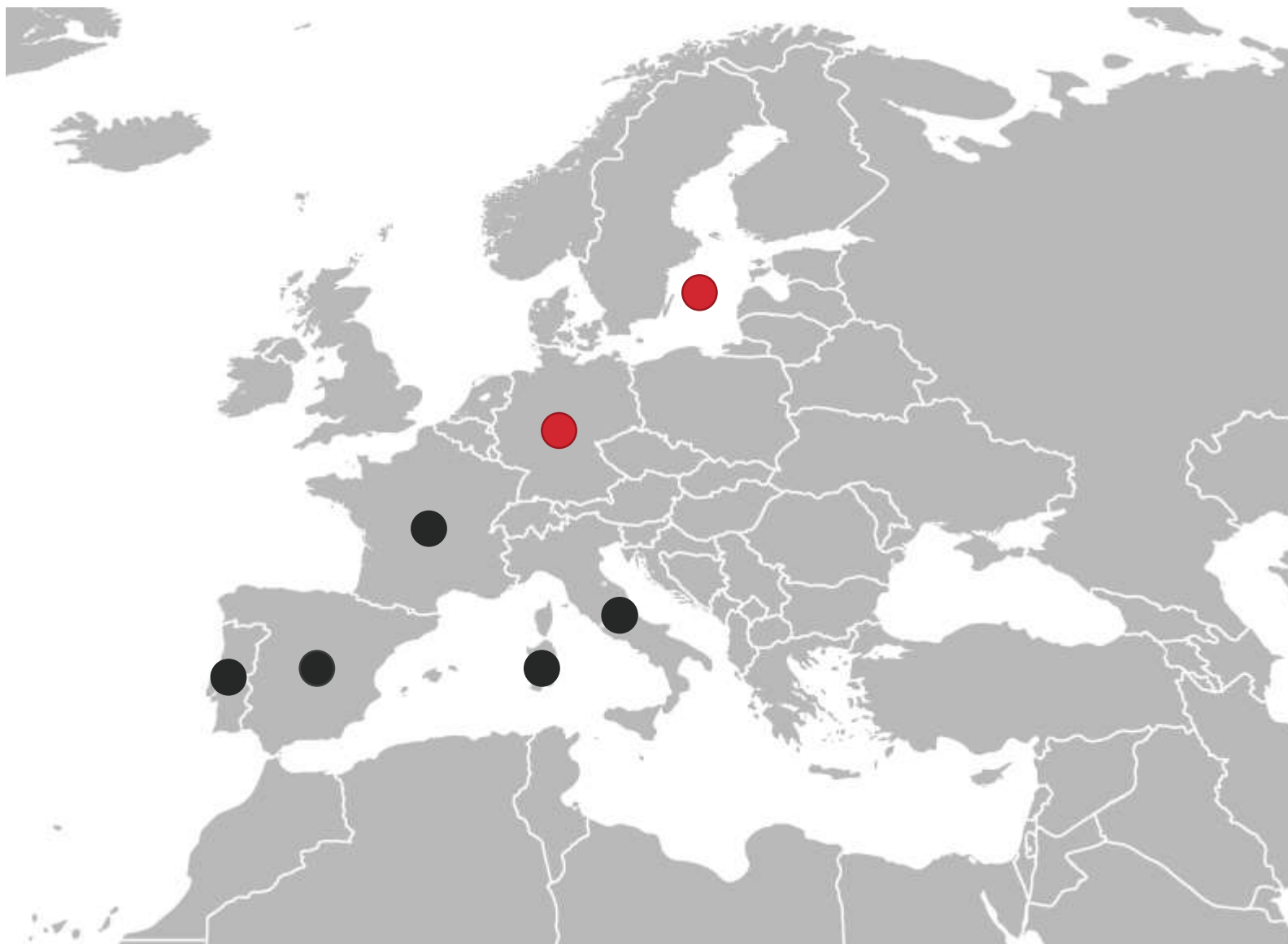
2011



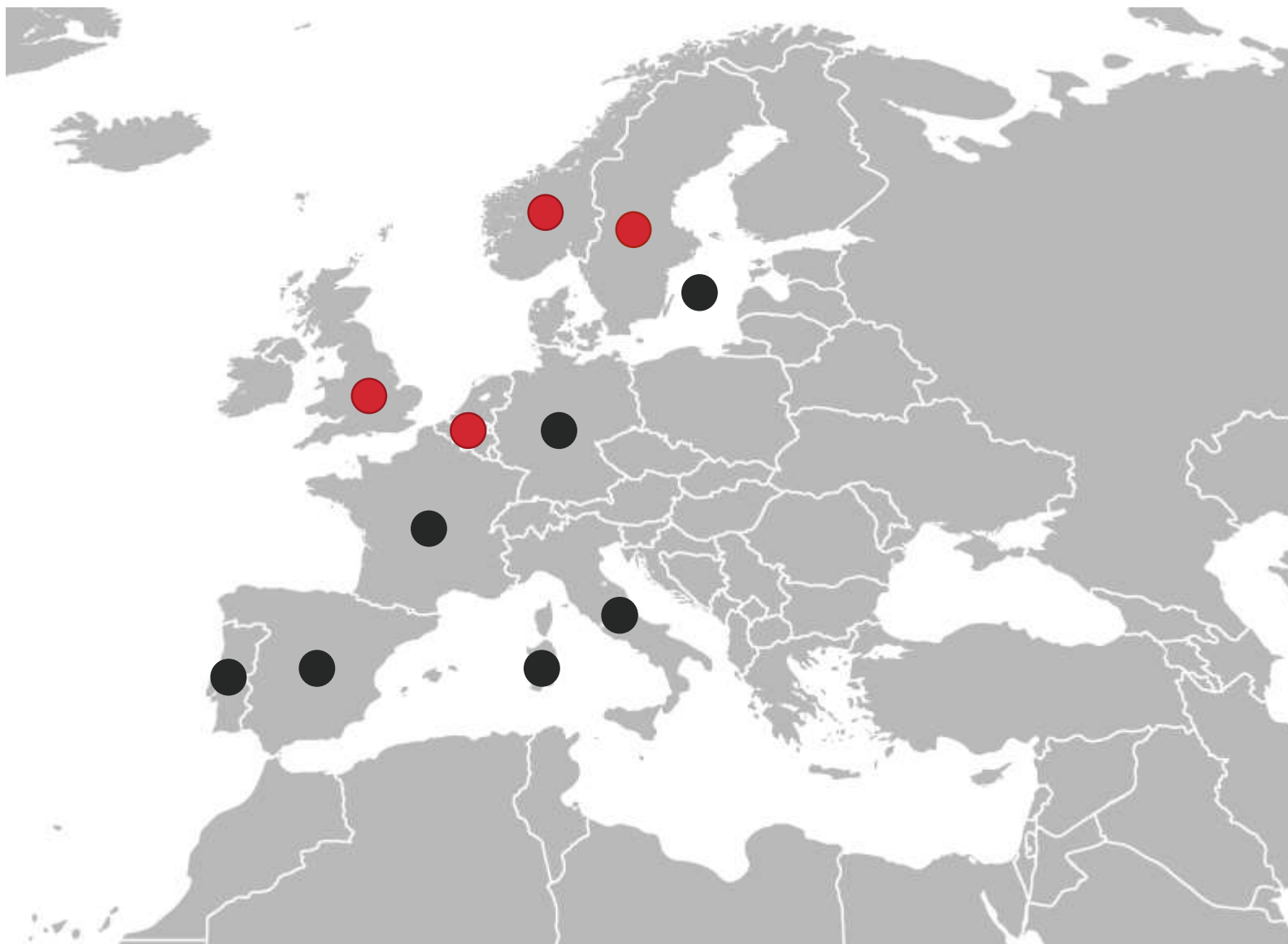
2012



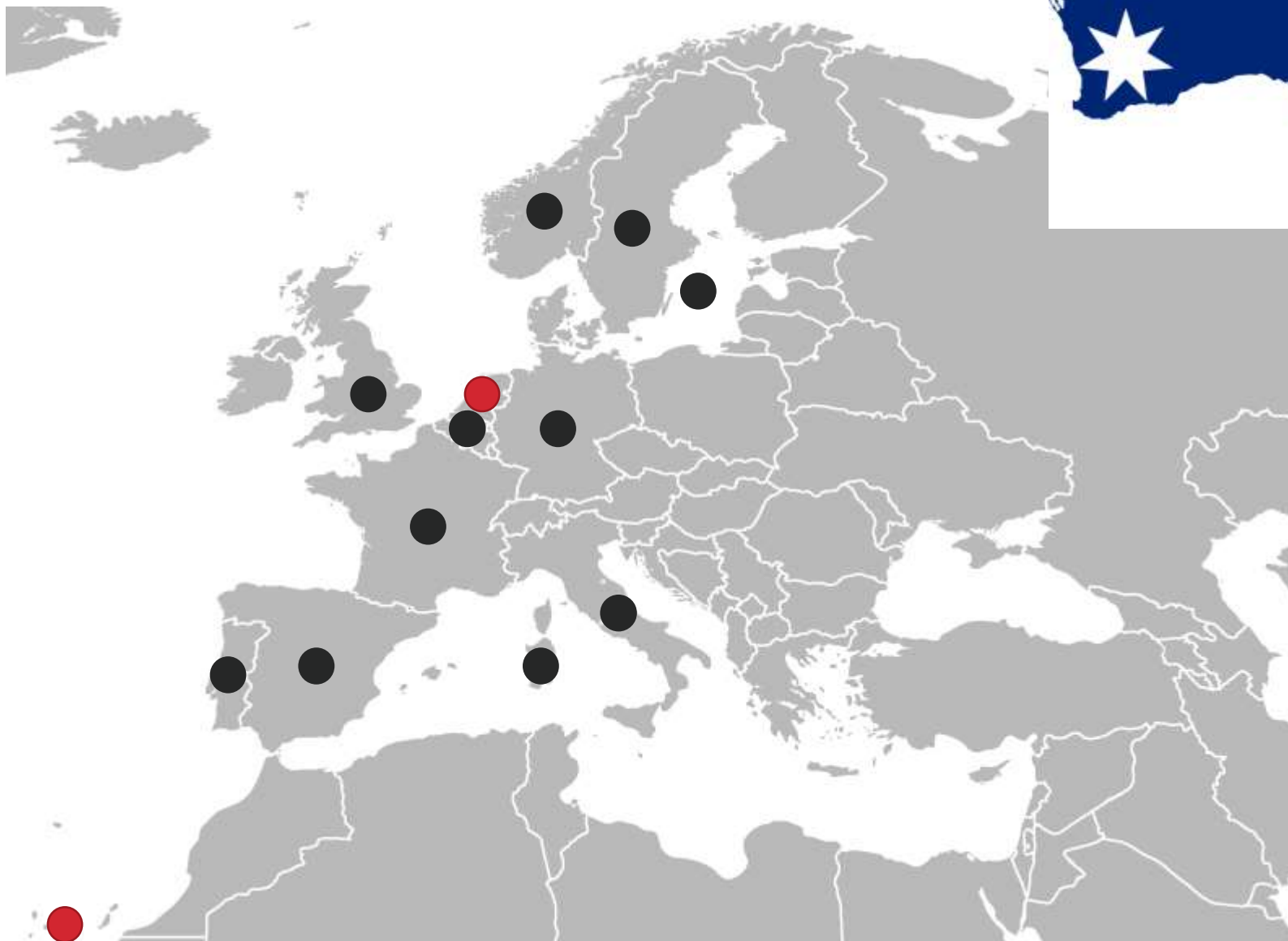
2013



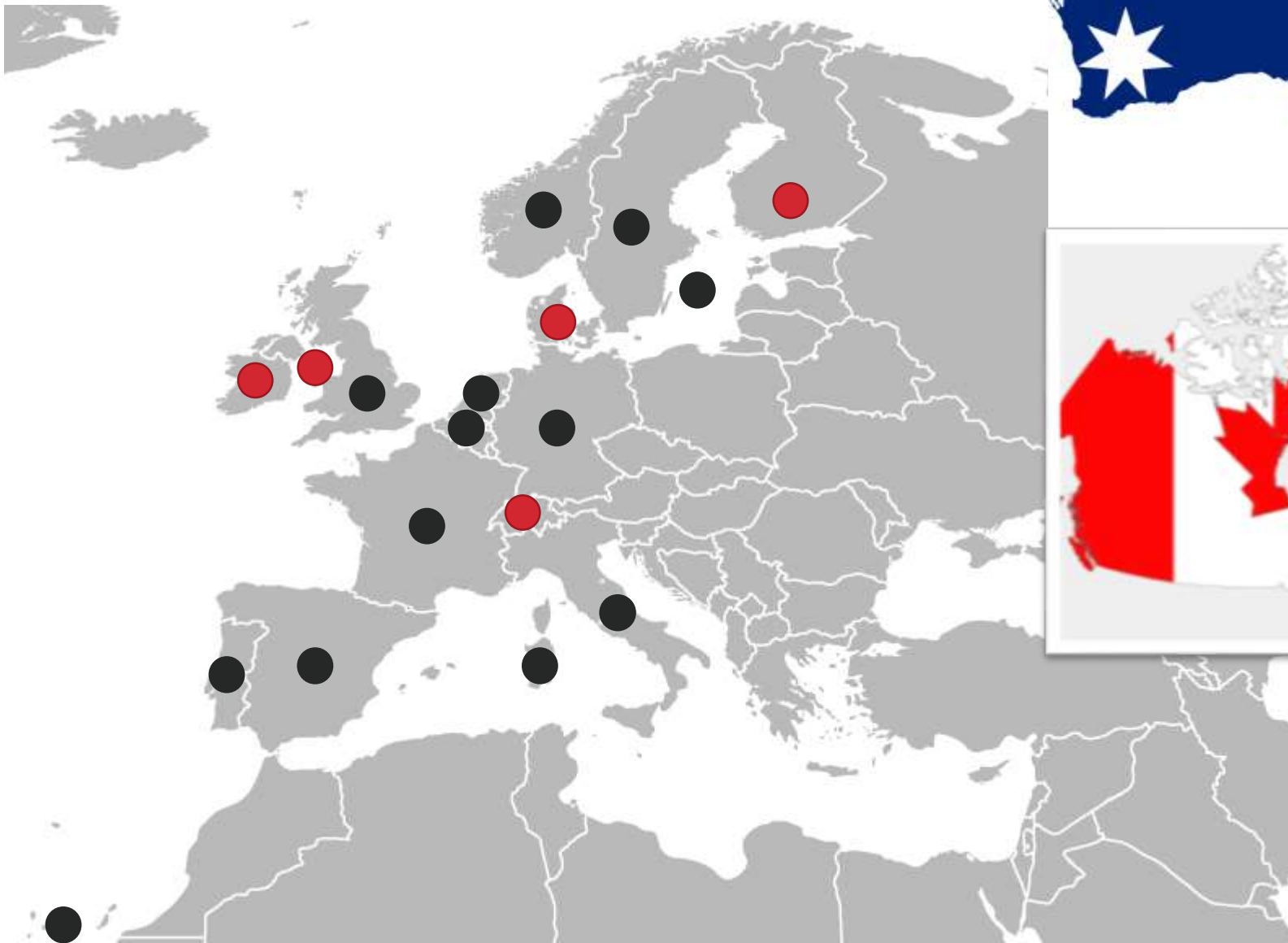
2014



2015



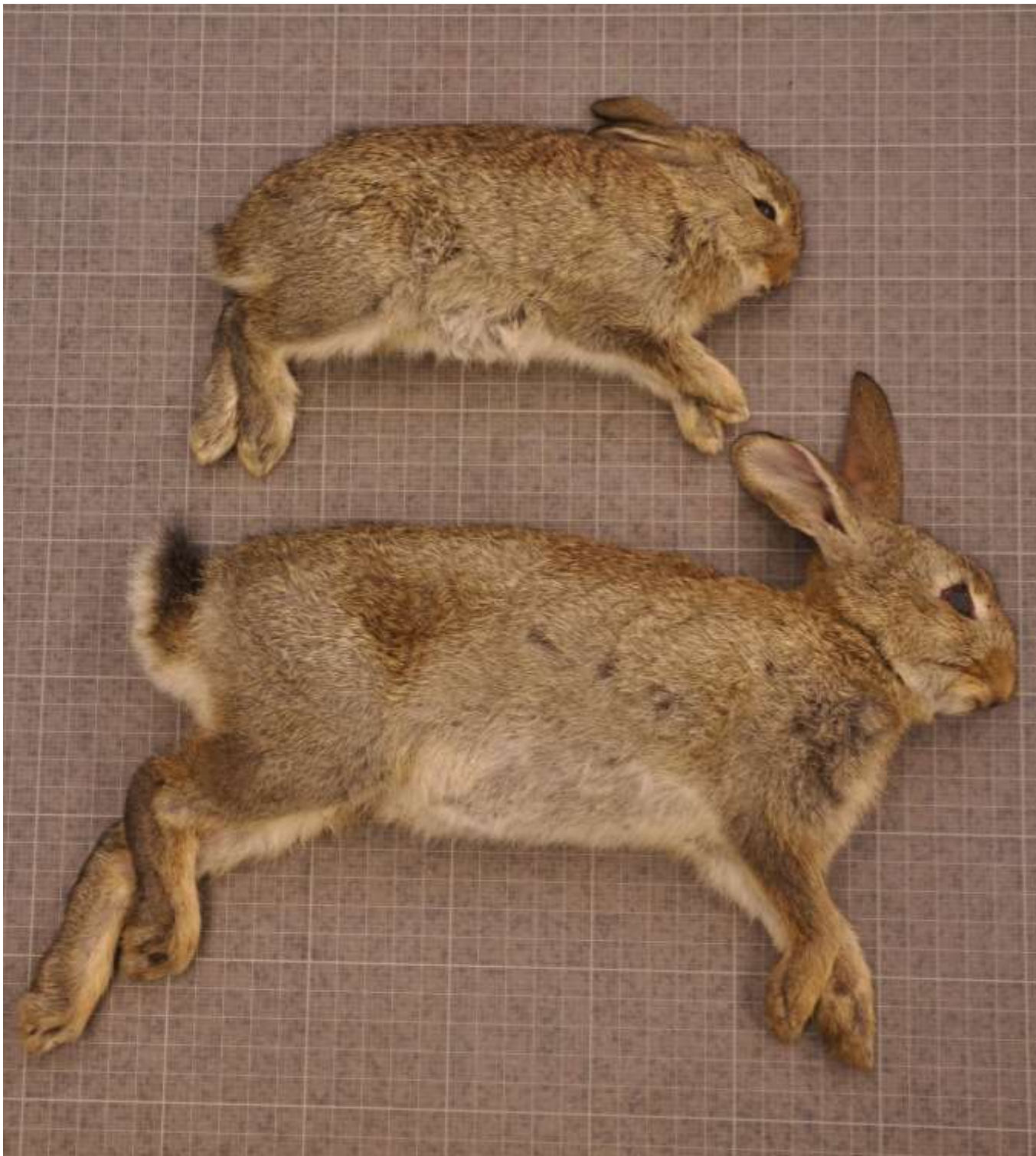
2016





IMMUNOLOGICAL BARRIERS

- Antigenically distinct from RHDV/RHDVa
- Immunity against RHDV partially protective
- Problem for domestic rabbit owners and producers- practical barrier of access to RHDV2 vaccines



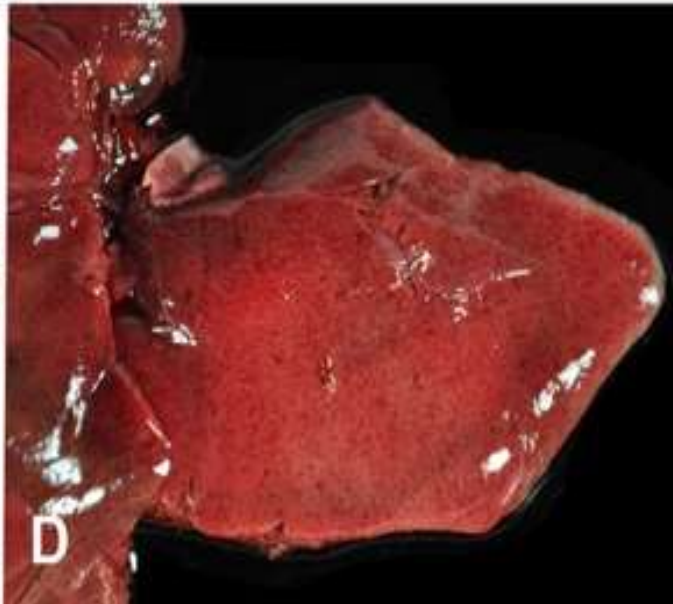
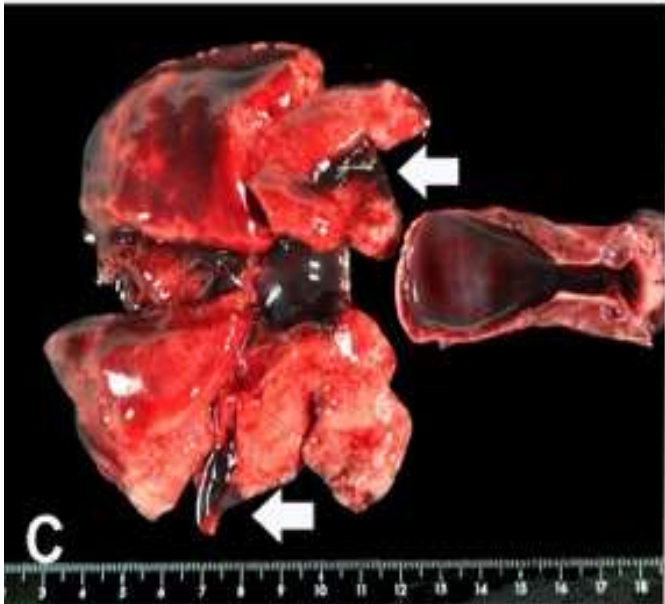
EPIDEMIOLOGIC BARRIERS: AGE

- RHDV only causes clinical disease in rabbits from 5-8 weeks of age
 - Mechanism not completely understood: weak binding to HBGA, innate immunity also involved
- RHDV2 infects and kills rabbits younger than 4 weeks of age
- Potential impacts on epidemiology of RHD in wild rabbit populations



EPIDEMIOLOGIC BARRIERS: HOST SPECIES

- RHDV and the related European Brown Hare Syndrome Virus are host-specific



EPIDEMIOLOGIC BARRIERS: HOST SPECIES

- Cape hare (*Lepus capensis subsp. mediterraneus*)
- Italian hare (*Lepus corsicanus*)
- European brown hare (*Lepus europaeus*)

Velarde et al (2016) Transboundary and Emerging Diseases



K. Bernodt



H. Cedervind

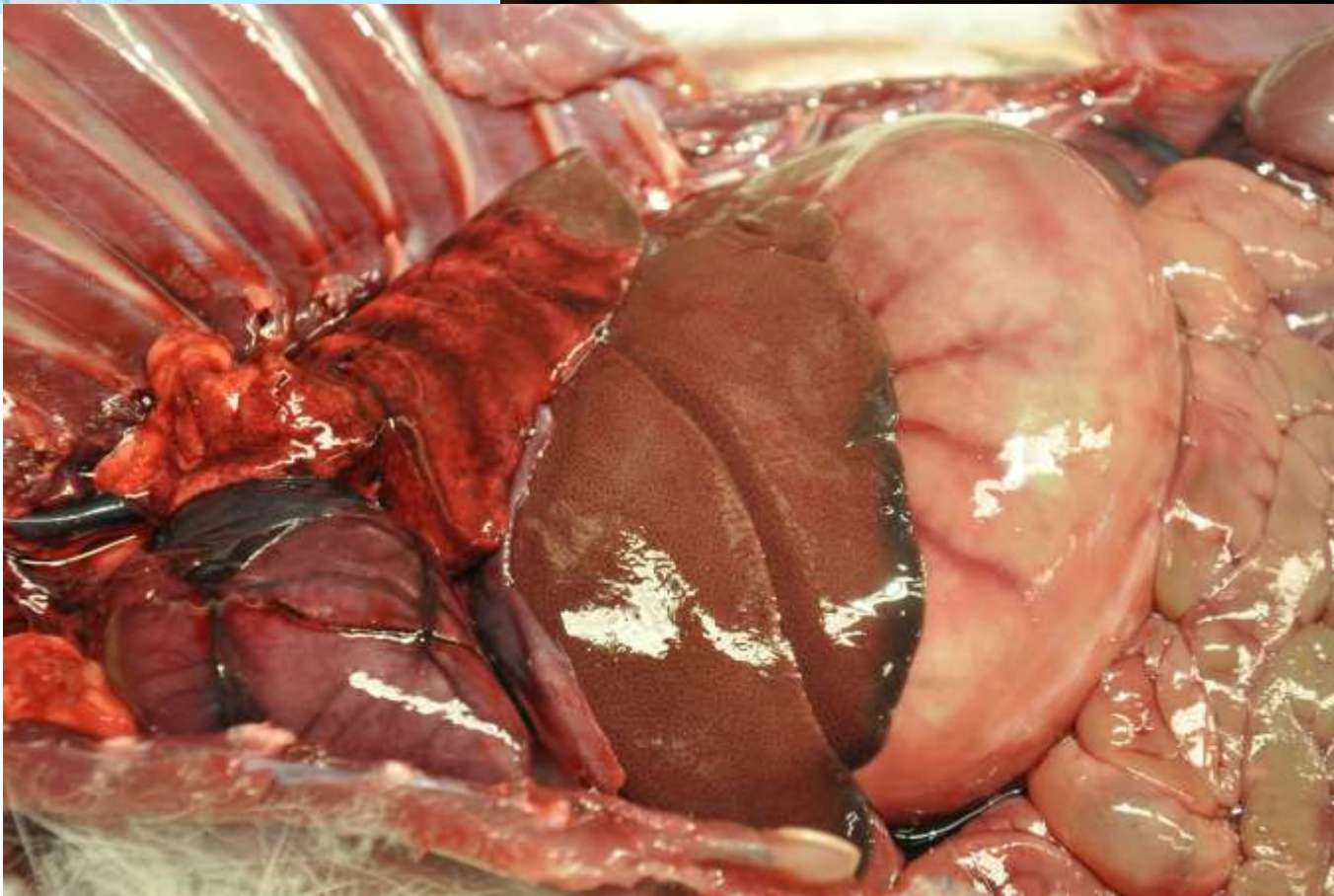
EPIDEMIOLOGIC BARRIERS: HOST SPECIES

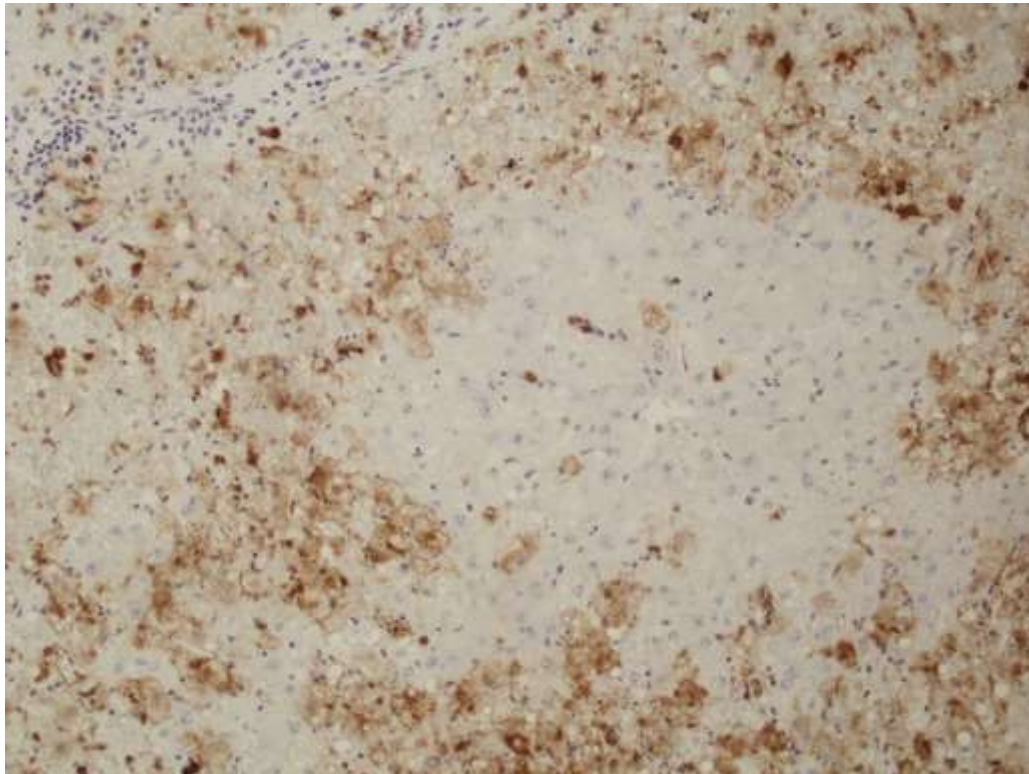
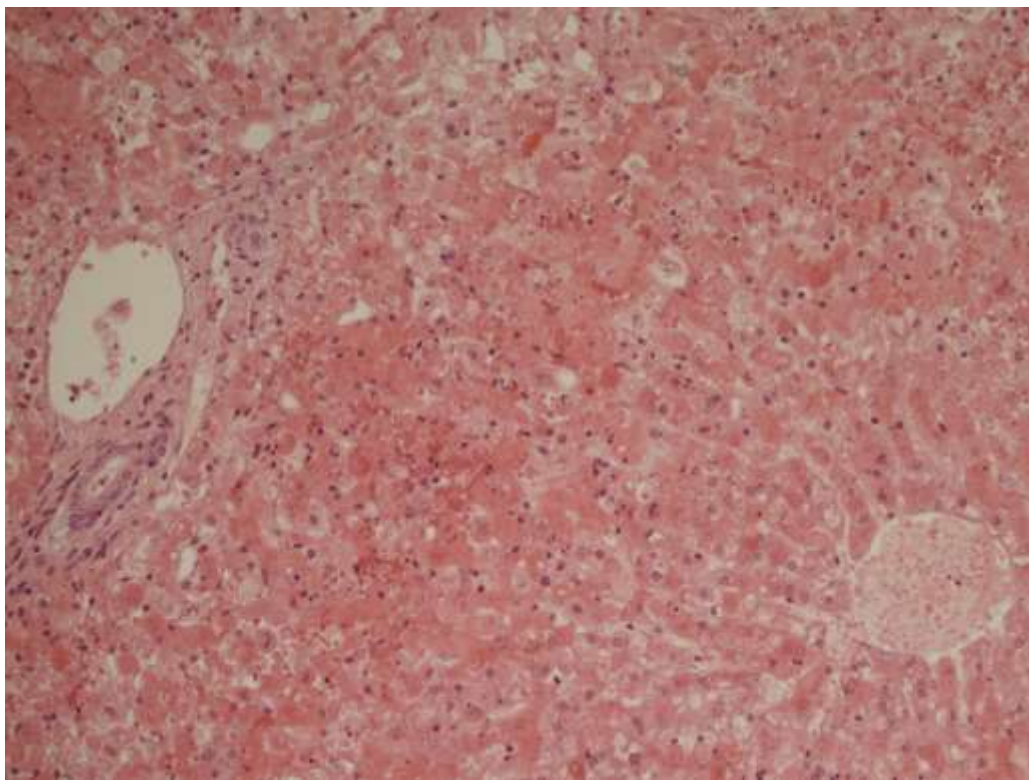
- Sweden has both Mountain hares (*Lepus timidus*) and European Brown Hares
- *Lepus timidus* subsp *sylvaticus* in southern Sweden
- Mountain hares declining in the southern half of Sweden where their range overlaps with brown hares
 - Interspecies competition with European brown hares
 - EBHSV



MOUNTAIN HARES IN SWEDEN

- In autumn 2016, mountain hare die-off on a small island off of the south-west coast of Sweden
- Three carcasses submitted in November 2016, December 2016 and March 2017
- Classic lesions of EBHS
- RHDV2 virus detected in all three animals
 - Island has no population of the natural host (wild rabbits)
 - Only 3 km from adjacent mainland
 - 50 000 tourists visit the island every summer

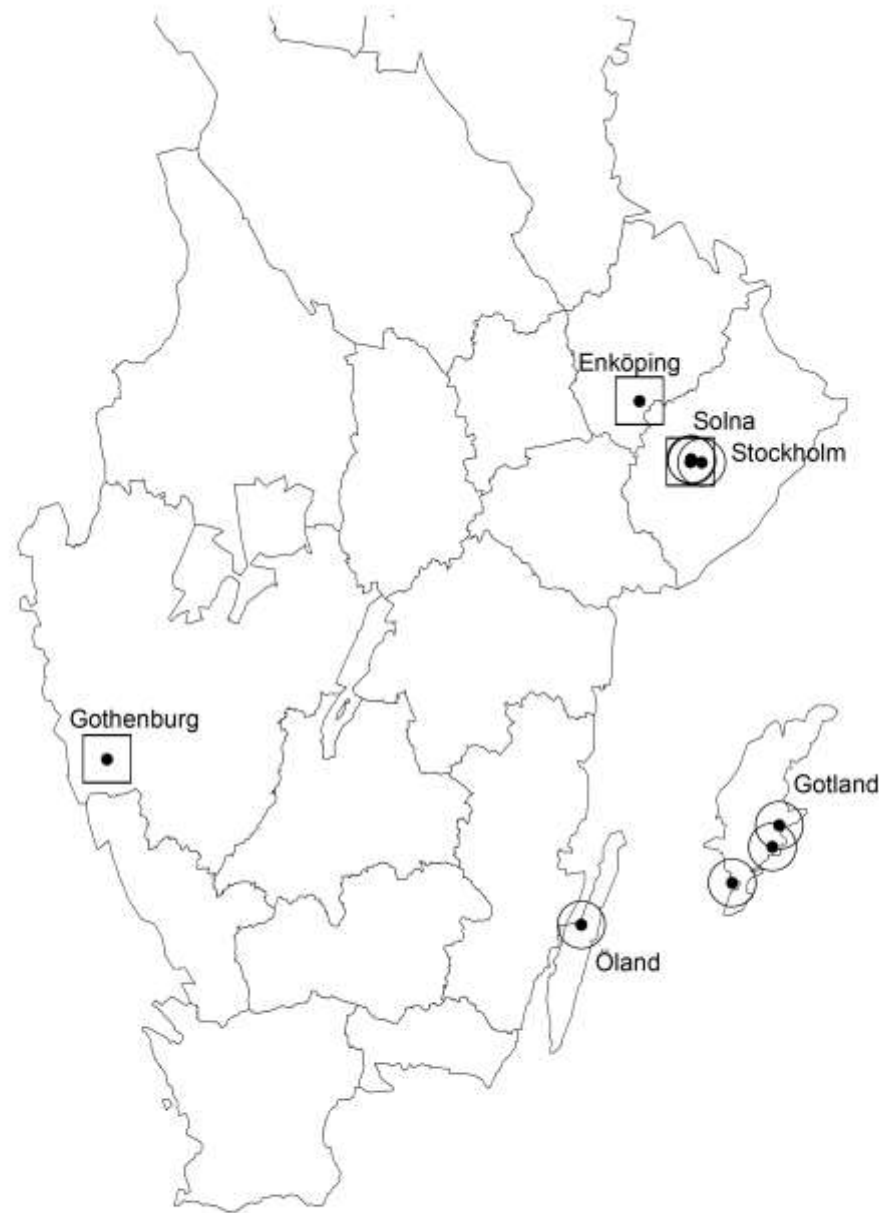




ANALYTICAL BARRIERS

- Analyses for confirmation of RHD in Sweden include nested PCR (Ros Bascuñana et al 1997) and immunohistochemistry for lagoviruses
- Diagnostic tests for RHD did not discriminate between RHDV and RHDV2

RHDV2 outbreaks in Sweden May 2013- May 2016

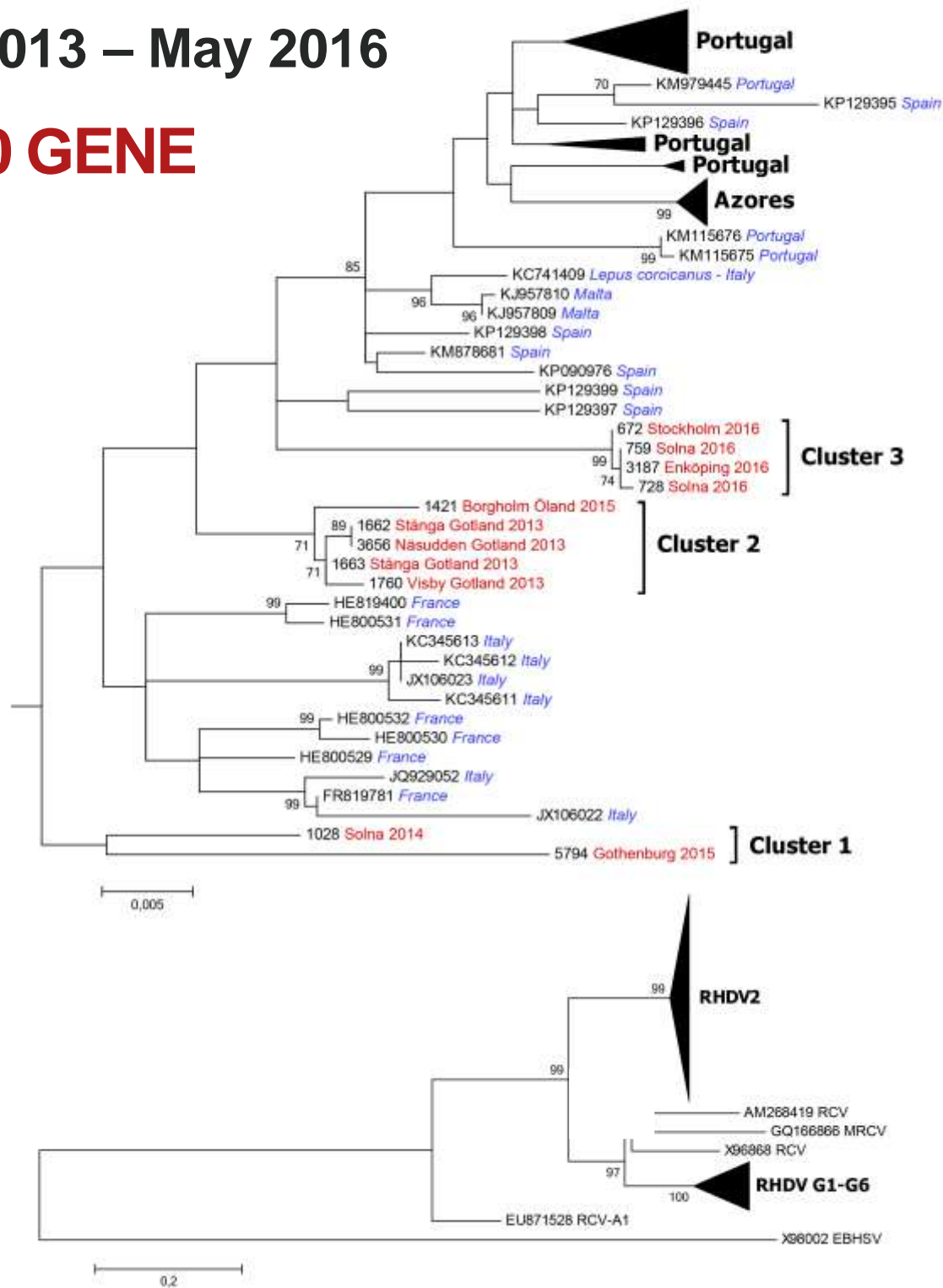


ANALYTICAL BARRIERS

- Presence went undetected in Sweden and likely elsewhere
- Development of new analytical methods
- RHDV2-specific RT-qPCR targeting the VP60 gene (Duarte et al 2015)

May 2013 – May 2016

VP60 GENE



- Swedish strains cluster according to time and geographic location
- Viral evolution, multiple introductions or both



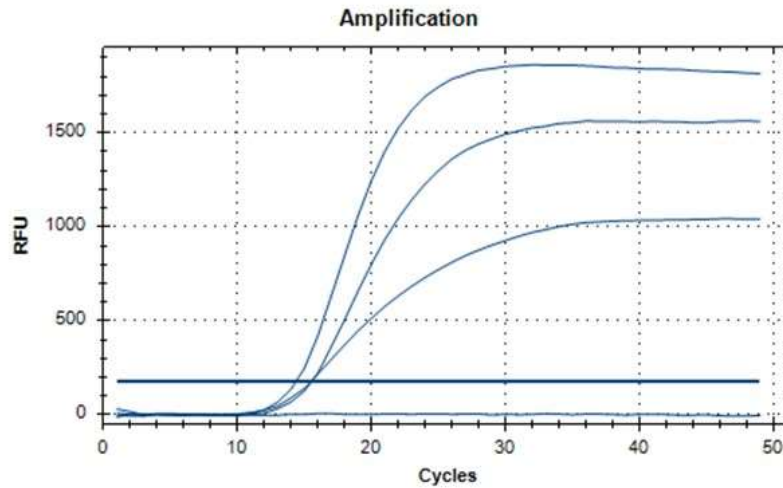
A RAPIDLY EVOLVING VIRUS

- Classic RHDV: acute disease, 80-90% mortality
- Early RHDV2 strains: subacute disease seen more often, 20-30% mortality
- More recent RHDV2 strains: acute disease, 80% mortality

Capucci et al Vet Rec 2017

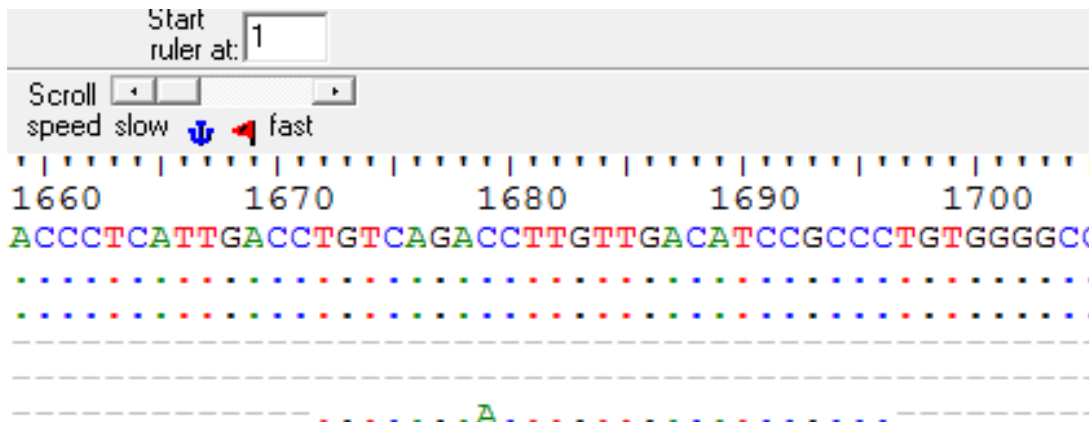
Sample Cq

2016 15,42
2021 15,48
1120 14,33
NTC



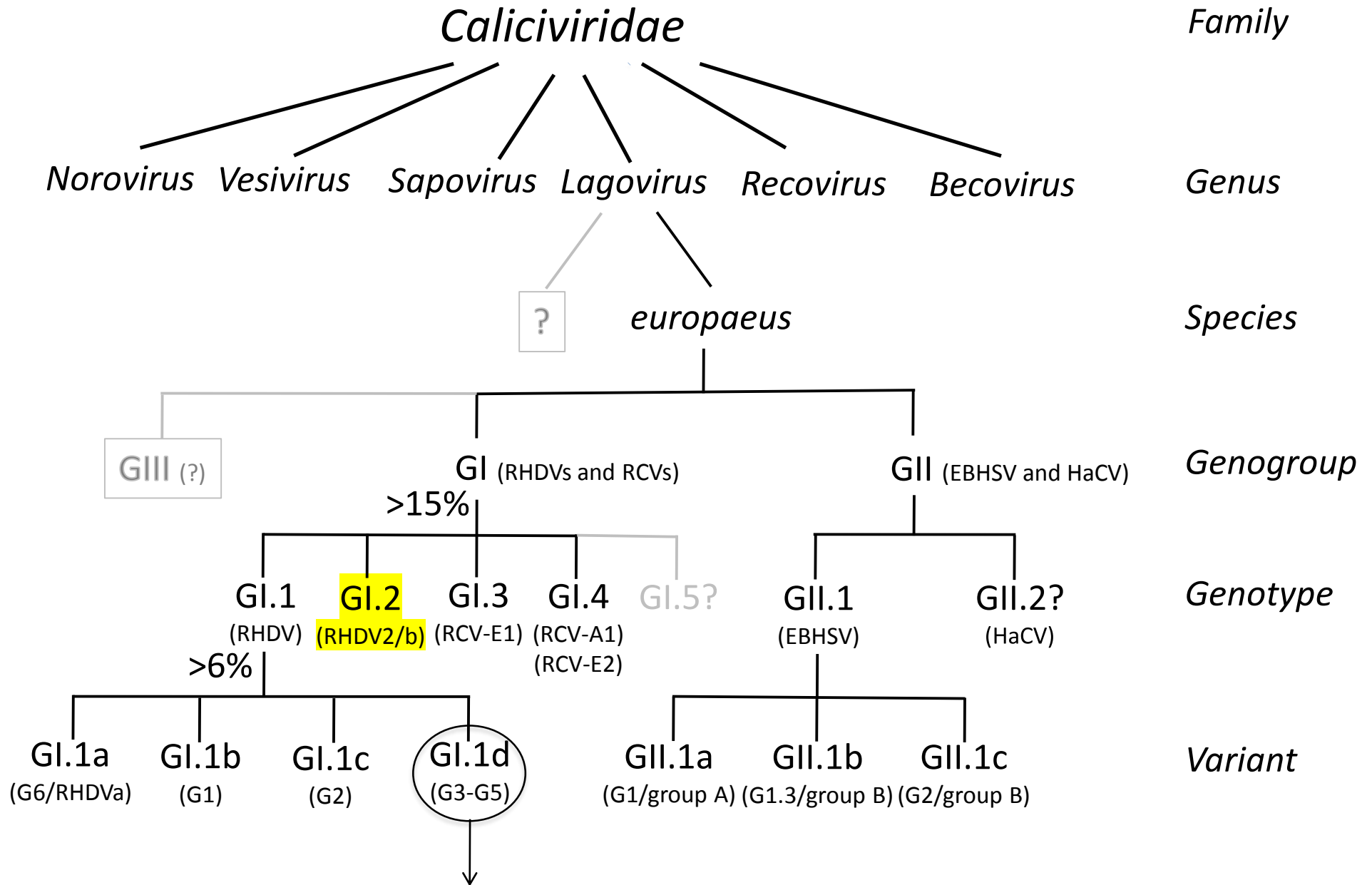
ANALYTICAL BARRIERS

- Rapidly evolving virus
- Efficacy of analytical methods must be regularly monitored



THE NOMENCLATURE BARRIER

- Le Pendu et al., Journal of General Virology 2017;98:1658–1666
- Blurred host species boundaries, new lagoviruses of varying pathogenicity, recombination
- Based on phylogenetic relationships
- Single species: *Lagovirus europaeus*, then divided into genogroups, genotypes and variants based on genetic distances



Comprehensive name: *Lagovirus europaeus/GI.1d/O cun/FR/2003/03-24*

Common name: *GI.1d_RHDV_03-24*

CONCLUSIONS

- While RHDV2 is similar to classic RHDV, it has crossed two notable barriers: age range and host range
- Mechanisms responsible for these changes and impacts of these changes on disease dynamics in the field warrant further investigation
- RHDV2 continues to evolve, necessitating regular re-evaluation of diagnostic tools and vaccine efficacy
- Origins of RHDV2 and other pathogenic lagoviruses are unresolved

ACKNOWLEDGEMENTS

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- **Swedish Environmental Protection Agency**



Erik Ågren

EPIDEMIOLOGIC BARRIERS



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ww



del Linca

- What is the role of other species?
- Viral RNA in sympatric wild rodents
- RHDV2 as a 'keystone species' on the Iberian peninsula