

Sudden increase in seroprevalence of Dobrava-Belgrade virus (DOBV) in *Apodemus flavicollis* in the province of Trento, Italy

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Abstract: Dobrava-Belgrade virus (DOBV) is considered the most pathogenic of hantaviruses in Europe, causing HFRS with case fatality rate up to 12%. The first DOBV strain was isolated from lungs of *A. flavicollis* mice captured in Slovenia, southeast Europe, and now represents the prototype DOBV strain (named Slovenia, or Slo/Af) from *A. flavicollis*. The spatial and temporal distribution of hantaviruses has been monitored in Trentino region (northern Italy) since the year 2000 by performing seroprevalence studies in rodents and humans with IFAT. An intense long term serological monitoring of a population of *Apodemus flavicollis* has been carried out. During the period 2000 – 2008 the antibody seroprevalence of DOBV varied from 0% to 1.4% . From the year 2010 onwards a sudden increase has been registered: 3,8% in 2010, 3% in 2011 and 7,7% in 2012. Possible drivers affecting changes in DOBV seroprevalence in this rodent population will be discussed.

Introduction:

The rapid emergence and geographic expansion of hantaviruses that cause HFRS (Haemorrhagic Fever with Renal Syndrome) in Europe are of particular concern for public health in Europe (Vaheri et al., 2012) .

Local emergence is driven by the complex interaction of several factors which include the rodent host population density, structure and temporal dynamics, behaviour, habitat vegetation coverage and structure, snow cover and food supply, host genetics, virus shedding, virulence and persistence in the environment (Jonsson et al, 2010; Reusken and Heyman, 2013). Several of these factors are still poorly known and obtaining quantitative parameters is a challenge.

Dobrava-Belgrade virus (DOBV) is a human pathogen that has evolved in, and is hosted by, mice of several species of the genus *Apodemus*. DOBV was first isolated from a yellow-necked mouse (*Apodemus flavicollis*) captured in a natural focus of HFRS in Dobrava village, Slovenia (Avsic-Zupanc et al, 1992). The species Dobrava-Belgrade virus are now classified into four related genotypes – Dobrava, Kurkino, Saaremaa, and Sochi – that show characteristic differences in their phylogeny, specific host reservoirs, geographical distribution, and pathogenicity for humans (Klempa et al., 2012).

Hantavirus antibodies in rodents and humans have been reported after cross-sectional epidemiological studies carried out in Trentino- Alto Adige region in northern Italy and part of the Austrian territory since the year 2004 (Kallio- Kokko et al. 2005, G. Walder et al., 2006). However, despite the evidence of human exposure to hantavirus in these regions, the number of confirmed human cases in Italy remains to be assessed (Heyman et al., 2011).

Within our research of wildlife zoonotic diseases in Trentino region, we present the preliminary results of our long term hantavirus serological monitoring of a population of yellow-necked mouse (*Apodemus flavicollis*).

Materials and methods:

IFAT

Rodent sera were screened using Immunofluorescent antibody test (IFAT) against Dobrava/Saaremaa virus antibodies as described earlier (Kallio-Kokko et al. 2006).

Statistical analysis

To assess if hantavirus occurrence varies between the 2000-2009 and 2010-2012 periods a Generalized Linear Model (GLM) with binomial error distribution was implemented. To investigate differences between the same periods for climatic variables (minimum, mean and maximum temperature and rainfall) a Linear Model (LM) was used. We finally implemented a GLM with binomial error distribution to assess how hantavirus occurrence is affected by climatic variables, population rodent density, sampling grid, rodent weight, sex and larval tick burden. Starting from the full model a model selection was performed by computing all possible sub-models and ranking them in according to their AIC. For parsimonious best models ($\Delta AIC < 4$) a model averaging procedure was carried out following Burnham and Anderson (2002) approach.

Results

2077 rodent individuals were captured from 2000 to 2012 and sera samples collected and analyzed. For the whole period, the average hantavirus seroprevalence was 1.73% (s.e.=0.29%), ranging from 0 (in 2000, 2002 and 2003) to 12% (in 2012). The prevalence observed during the period 2000-2009 (0.71% \pm 0.20%) was lower than in the following period 2010-2012 (6.9% \pm 1.42%), (z-value=5.80, $p < 0.001$), (Fig. xx, panel b). For climatic variables only the yearly average daily rainfall was different between 2000-2009 and 2010-2012 periods with higher precipitation during 2010-2012 (t-value=2.02, $p < 0.05$), (see Fig. xy, panel d). The output of the model averaging procedure for hantavirus occurrence during all the period considered (from 2000 to 2012) is reported in Tab. xx, where coefficient estimates and statistics of selected explanatory variables are reported, and in Fig. xz, where model weights of selected explanatory variables are shown.

Discussion and conclusion

It has been shown that the emergence and spill over of several zoonotic pathogens is usually characterized by a long silent latent circulation within his wildlife reservoir host followed by a sudden rise as consequence of changes in one or more of the driving factors which basically include intrinsic pathogen and population factors and extrinsic environmental conditions which affect pathogen persistence in the environment as well as host population dynamics (Mills, 2005; Mills, 2006). Our results show that DOBV circulated silently for a long time within a population of *Apodemus flavicollis* as we could observe until 2009 and then showed a sudden rise. Because of the chronic features of the hantavirus infections, factors affecting contact rate among individuals and persistence of the virus in the environment should be carefully examined. In fact, indirect transmission makes the transmission rate less dependent on the host density and enhances the persistence of virus in the host population (Sauvage et al., 2003).

Our observations and analysis showed that:

- A rise in precipitation and temperature (especially the T max) has been observed also at local small scale
- Population density do not explain alone the observed variation in prevalence

Work in progress:

We are currently progressing our analysis to obtain virus isolation and molecular typing and phylogenesis, to study variation in MHC genes and better explore possible variation in rodent behaviour and population dynamics.

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