



RAPID RISK ASSESSMENT

New bornavirus strain detected in the EU

25 February 2015

Main conclusions and options for action

A recently reported cluster of acute fatal encephalitis in three squirrel breeders possibly related to an infection with a newly identified bornavirus is an unusual event. The novel nature of this occurrence requires that additional investigations are undertaken into the role of a new bornavirus in the aetiology of these cases, the identification of natural hosts, reservoir and the transmission route.

Nevertheless, pending the completion of the cluster investigation, it is advised that feeding or direct contact with living or dead variegated squirrels should be avoided, as a precautionary measure.

Further investigations are ongoing to characterise these cases. In addition, testing of cases of human encephalitis for this newly identified bornavirus, especially in areas where the presence of bornavirus is documented in animals, can contribute to a better understanding of the risk of bornavirus infection in humans.

Source and date of request

ECDC internal decision, 20 February 2015.

Public health issue

A new bornavirus detected in a variegated squirrel potentially associated with three fatal encephalitis cases in squirrel breeders in Germany raises the question of whether this represents an emerging threat in the EU.

Consulted experts

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Disease background information

Bornaviruses are enveloped, 80 to 100 nm in diameter, negative sense RNA viruses of the Bornaviridae family [1]. The 8.9 kilobases viral genome encodes for six proteins, among which are the nucleoprotein (p40, N) and the polymerase cofactor phosphoprotein (p24, P) [2,3]. Replication of the bornavirus genome occurs inside the nucleus of host cells. Close association of the virus with chromosomes facilitates virus spread from cell to cell during mitosis [4]. Bornaviruses show unique limited genome amplification that favours non-cytolytic virus persistence and evasion of the antiviral host response [5]. Integrations of bornavirus N-related sequences and sequences related to matrix and polymerase genes are reported in the genome of several mammalian species, such as in primates and the thirteen-lined ground squirrel (*Spermophilus tridecemlineatus*) [6,7]. Bornavirus-like genes have also been reported in reptiles, indicating a wide host range [8,9]. Phylogenetic analysis of exogenous bornaviruses and mammalian endogenous bornavirus-like (EBLs) elements demonstrates that bornaviruses have co-existed with primates in evolutionary history for at least 40 million years [6].

Borna disease in animals

Borna disease virus (BDV) infections were first described in the 18th century and were named after the town of Borna, near Leipzig, Germany, where an epizootic condition was described in 1885 among military horses presenting with a fatal neurologic disease [10]. BDV can infect many vertebrate species including rhesus monkeys, horses, sheep, cattle, goats, rabbits, deer, llamas, alpacas, cats, rats, mice, shrews, gerbils, dogs, and ostriches. BDV related avian bornaviruses (ABV) have been recently described in psittacine birds [11], Canada geese, trumpeter and mute swans [12], and canary birds [13,14].

Animal BDV infections have been described in central Europe, the United Kingdom, Israel, Japan, Sweden, Finland, Australia, and the United States of America [15]. In 2000 in Austria, Nowotny et al. [16] described a new subtype of BDV isolated from a horse that was euthanized due to severe and incurable neurological disease. The new subtype differed from the reference strains by more than 15% and was difficult to detect by standard reverse transcriptase PCR protocols. This indicated that the genome of BDV is far more variable than previously assumed and that naturally occurring subtypes may escape detection by currently used diagnostic assays.

In animals, BDV infection may lead to an acute or sub-acute disease with meningo-encephalitis or mild manifestations with alteration or impairment of nerve-cell functions [10]. The specific disease syndromes depend on many host factors including the species, breed, age, and immunological status of the animal at the time of infection. In classical Borna disease, paralysis is common and death occurs within five weeks in the majority of animals. Recovery is possible with life-long altered behaviour [13]. Avian bornaviruses are the causative agents of pro-ventricular dilatation disease, a fatal neurological disease considered to be a major threat to psittacine bird populations [9].

Small wild animals are the most probable vectors and reservoirs of BDV infection, although the epidemiology is not fully understood. Arthropods and birds have also been investigated as possible BDV reservoirs [11]. Experimental animals are most efficiently infected with BDV via intranasal or intracerebral routes, and via intramuscular, intradermal or subcutaneous routes, but not intragastrically [17]. Infection may occur via excretions e.g. urine or faeces. As with rabies, BDV spreads intra-axonally from the inoculation site into the central nervous system [17].

Bornaviruses in humans

The first evidence of possible human infections with bornaviruses resulted from a study published in 1985 showing that patients with major depressive psychiatric disorders had a higher bornavirus-related seroprevalence as compared with the control group [18]. The majority of later studies also reported the higher seroprevalence or other BDV markers in patients presenting with diverse psychiatric disorders [19]. However, it was not possible to infer the causal relationship between BDV infection and psychiatric disorder because of the occasional presence of the infectious markers in control patient groups, proven viral contamination of laboratory samples and the disagreement on diagnostic methods for BDV. However, very few studies that employed a combination of reproducible and controlled methods suggest that BDV infection in humans is possible [20-22]. Nevertheless, the frequency of human infections, the role of endogenised bornavirus genes, and the existence of potential 'human BD' are still uncertain [10].

Event background information

On 19 February 2015 Germany posted a message on EWRS, reporting three cases of fatal encephalitis in residents of the state of Saxony-Anhalt. The first clinical case was seen in 2011, and the second and the third in 2013 in different hospitals. Affected persons were males aged 62 to 72 years and of age-typical health status. Each of them was known to breed variegated squirrels (*Sciurus variegatoides*), a type of tree squirrel common to Central America that can be kept as an exotic outside pet. They knew each other but did not live in close proximity to one another. It is unclear whether they exchanged animals. During the prodromal phase, which lasted for two weeks or longer, the patients presented with fever and shivering, fatigue, weakness and walking difficulties. Due to increased confusion and psychomotor impairment they were admitted to neurology wards where they developed ocular paresis. They rapidly deteriorated within a few days and died after some time in intensive care, despite mechanical ventilation. Investigations for usual (non-purulent) encephalitis aetiologies performed at the Bernhard Nocht Institute for Tropical Medicine in Hamburg at first did not produce any evidence for known pathogens in cerebrospinal fluid and samples of brain tissue of the deceased.

The Friedrich Loeffler Institute on Riems Island investigated the carcass of one variegated squirrel belonging to the third patient. Genetic analysis of a tissue sample pool of the animal using a metagenomics approach produced sequences of a newly identified type of bornavirus. Further molecular and immunohistochemical analysis of brain tissue from the three deceased patients confirmed presence of this virus in the human cases as well. The newly identified virus is clearly different from all currently known bornaviruses.

Tissue and cerebrospinal fluid collections from bio banks are now being investigated for the identification of additional cases. Previously unresolved cases of encephalitis are being re-evaluated in view of the new virus. Breeders/owners of variegated squirrels will be questioned as to their health condition and asked about anecdotal knowledge of possible fatal cases among breeders/owners. The available evidence is compatible with this virus being a zoonotic pathogen. In limited testing of additional variegated squirrels from one breeder and a zoo no other animal was found to be positive for this infection, but further tests are ongoing. Breeders are asked to send in deceased animals to the Friedrich Loeffler Institute.

ECDC threat assessment for the EU

Several findings support the presumption that the three cases of encephalitis in humans are associated with a newly identified bornavirus that was very likely to have been transmitted from infected squirrels to humans:

- Genetic analysis (metagenomics) of brain tissue of a variegated squirrel produced sequences of a newly identified type of bornavirus with less than 77% homology with viruses of the Mammalian 1 cluster
- Using the same methodological approach the same gene sequences were identified in brain samples of three squirrel breeders who presented the same clinical picture of fatal encephalitis
- All human cases were squirrel breeders
- The metagenomics approach has previously been validated as a powerful tool for investigation of unknown pathogens, as demonstrated when used for Schmallenberg virus identification.

However, the cases are still under investigation, and proof of a direct causal relationship between the presence of genetic material in human brain and encephalitis has not been established. Additionally, the presence of genetic material of bornavirus can result from endogenisation of ancient bornavirus genes in vertebrate genomes.

Variegated squirrels appear to have been introduced only recently in Europe. The number of animals and breeding places in Europe is unknown. The squirrels originated from Central America, and could have been imported with the infection or they could have become infected in Germany through contact with other infected animals. The current investigations being undertaken by the Robert Koch Institute with the local authorities has been extended to other wild species in the region. Breeders, often previous bird keepers, usually keep squirrels in a stable outside in their backyard. The Magdeburg Zoo in Germany seems to be the only zoo in Europe to hold a variegated squirrel pair (<http://www.zootierliste.de/en/?org=7>).

Data on the distribution of the virus in the variegated squirrel population are not available and it is not clear whether squirrels or other animals/ arthropods are reservoirs or vectors. It is unclear if other mammals could be infected with this newly identified bornavirus.

The transmission route in these three cases has not been determined [23], although it is plausible that people may become infected, probably through bites or scratches. Airborne transmission as observed for Hantaviruses cannot be excluded for this bornavirus infection, e.g. via inhalation of air contaminated by virus from faeces or urine of infected animals.

As exposure or contact with the squirrels that are the suspected vector/reservoir appears not to be common, and other cases among contacts of these cases have not as yet been identified, the risk for the general population is considered to be very low. Furthermore, the possibility of zoonotic infection with a newly identified bornavirus has not been proven. Nonetheless, squirrel breeders, their family members and the owners of squirrel pets could be at higher risk of exposure to infection. However, the risk of disease transmission from squirrels to humans, and human to human transmission cannot be assessed because of the low number of cases reported, the lack of information about new virus biology and pathophysiology in humans, the lack of information about transmission and the prevalence of infection in squirrel population, and the absence of evidence on whether squirrels are a reservoir or a vector of the disease.

Conclusions

This cluster of acute fatal encephalitis in three squirrel breeders possibly related to an infection with a newly identified bornavirus is an unusual event. The role of new bornavirus in the aetiology of these cases, the identification of natural hosts, reservoirs and transmission route require additional investigations.

Nevertheless, pending the completion of the cluster investigation, feeding or direct contact with living or dead variegated squirrels should be avoided as a precautionary measure.

Further investigations are ongoing to characterise these cases. Testing cases of human encephalitis for this newly identified bornavirus, especially in areas where the presence of bornavirus is documented in animals, can contribute to a better understanding of the risk of bornavirus infection in humans.

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