

Recommendation of the ZKBS on the risk assessment of
Variegated squirrel 1 bornavirus
as donor or recipient organism according to Article 5 paragraph 1 GenTSV

General

The *Variegated squirrel 1 bornavirus* (VSBV-1) is a recently discovered member of the family *Bornaviridae* with zoonotic potential. The linear genome consists of a single-stranded RNA segment of negative polarity with a total length of approximately 8.8 kb [1]. The virus replicates in the nucleus.

Between the years 2011 and 2013, three breeders of *Variegated squirrels* in Saxony-Anhalt (Germany) successively contracted severe, progressive (meningo-) encephalitis with, in all cases, similar symptoms of central nervous system impairment (fever, chills, progressive psychomotor deceleration, confusion, unsteady gait, convulsions, ophthalmoplegia). All three patients also had bilateral deep vein thrombosis, which in two of the patients resulted in pulmonary embolism. The cause of the thrombosis is still unclear. Examinations indicated an unknown viral infection and the patients received broad anti-infective chemotherapy. They nevertheless went into a coma and died two to four months after the occurrence of the first clinical symptoms. All three patients were over 60 years old and had (age-typical) diseases such as hypertension, diabetes and obesity, which may have predisposed them to viral infection [1].

In a metagenome analysis, the Friedrich-Loeffler-Institut (FLI) was able to identify a matching gene sequence in a common *Variegated squirrel* (*Sciurus variegatoides*) derived from the breeders and in brain biopsies of all three deceased breeders, which indicated the occurrence of a novel bornavirus [1]. The genome was identified by *deep sequencing*. The annotation revealed the typical structure of a Bornavirus genome, with genes coding for the proteins N, X, P, M, G and L. Phylogenetic analyses have shown that this virus forms a lineage separated from the previously known Bornavirus species and that it is closest to the *Mammalian 1 bornavirus* (especially that of the horse) with a nucleotide sequence homology of 67.6% - 81.7%.

Bornaviruses of birds and reptiles have a minor genetic relationship to VSBV-1. Classical Bornaviruses have been known in Central Europe for decades to cause diseases with central nervous symptoms in various mammals. These lead to death in 90% of cases [2]. Whether these Bornaviruses lead to neuropsychiatric symptoms in humans, the so-called "human Borna disease", is a matter of controversy [3-6]. Due to the distant relationship with the Bornaviruses described so far, it was proposed to classify VSBV-1 as a new species [1].

In January 2016, the FLI announced that further infected animals could be identified in Germany in zoos and at private owners, among them also species of the *Beautiful squirrels* (*Callosciurinae*) [7; 8]. Whether VSBV-1 occurs in other animal species is not known. It is also

unclear whether the animals have already become infected in their native country Latin America or afterwards in Germany. The squirrels infected with VSBV-1 show no symptoms of disease.

High levels of VSBV-1 RNA were found in various organs of the squirrels, such as the brain, heart, lung, liver, kidney, spleen and colon, as well as in the *oropharynx* and in saliva. However, small amounts of viral RNA were detected in the blood and chest cavity fluid [1].

As already shown for other Bornaviruses, the virus could be released through various excretions from the infected squirrels [9]. Transmission between the squirrels could take place via the olfactory route, possibly via salivary and nasal secretions of infected animals, as suspected for the *Mammalian 1- and 2 bornavirus*; possibly also fecal-oral, as suspected for avian Bornaviruses [5]. A transmission by inhaling virus-containing dust is likewise not ruled out.

Zoonotic transmission to humans is most likely due to scratching and biting injuries [1; 7; 8]. Human-to-human transmissions are not known and not likely as the virus primarily infects the central nervous system in humans [1]. Since the cause of (meningo-)encephalitis remains unexplained in 40-70% of cases [10], it cannot be ruled out that further human VSBV-1 infections have already occurred worldwide.

The exotic squirrels are not endemic in Germany, but they are popular pets, which are often imported to Europe and other regions of the world. There is no specific treatment or vaccine available.

Recommendation

According to Article 5 paragraph 1 GenTSV in conjunction with the criteria in Annex I GenTSV *Variegated squirrel 1 bornavirus* (VSBV-1) is assigned to **risk group 3** as a donor and recipient organism for genetic engineering operations.

Reasoning

The *Variegated squirrel 1 bornavirus* infects *Variegated* and *Beautiful squirrels* and possibly other species. The infection remains asymptomatic in squirrels. VSBV-1 can be transmitted to humans and cause severe encephalitis, which was fatal in the three cases published so far. A human-to-human transmission has not been described previously. The route of transmission of VSBV-1 between the squirrels is unknown.

References

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