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West Nile Virus in Horses – An Update

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WNV was first isolated in December 1937, from a 37-year-old, febrile woman in the West Nile district in the Northern Province of Uganda. In Europe, human cases were documented in the 1960's. Currently, West Nile Virus is present on all continents. The West Nile virus belongs to the Flaviviridae family, like the dengue virus, the TBE virus and the Usutu virus. It is transmitted by various mosquitoes.

Number of cases

In Europe, most human and animal cases occur in Italy, Greece, Serbia and Romania. The cases reported to the European Centre for Disease Control (ECDC) for 2022 are shown in Table 1.

Table 1: WNV case numbers for the year 2022 *Source: ECDC*

	Cases Human	Deaths Human	Cases Horse/ Bird
Italy	586	37	47/258
Greece	284	31	9/–
Serbia	226	12	–
Romania	46	5	–
Germany	11	–	16/51
Austria	6	–	1/2
France	4	–	6/–
Spain	5	–	8/9
Hungary	14	–	3/1
Portugal	0	–	3/–
Croatia	8	–	–/2
Slovakia	1	–	–

Fig. 1: Culex mosquitoes are considered the most important vectors for WNV transmission

Image source: www.cdc.gov/mosquitoes

Pathogenic biology

WNV is a vector-borne virus of with wild birds being the main reservoir. These wild birds, often remain asymptomatic. However, corvids, passerine birds as well as certain birds of prey and owls can become severely ill and succumb to the virus. The virus is transmitted by ornithophilic mosquitoes, mainly *Culex* sp. (Figure 1). *Aedes* sp. and others such as the tiger mosquito can also be carriers. Humans and horses are considered dead-end hosts because the viral load during an infection is too low to infect other animals. Infections however, also been described in dogs, cats and sheep.

As a rule, arthropods (mosquitoes) are necessary for transmission. Individual cases of direct transmission through blood transfusion, organ transplantation, intrauterine transmission as well as person to person have been described.

Clinical image

WNV-associated diseases occur seasonally, from early summer into autumn, depending on the activity of the mosquitoes. As a rule, usually individual horses are affected. The regional distribution is also related to the flight routes of migratory birds (Figure 2).

In humans, about 80 % of infections remain asymptomatic, with febrile general illness (West Nile fever) developing in about 20 % of cases. Less than 1 % of human cases (1 in 150) develop WNV-associated meningoencephalitis, with severe infections, this is more prevalent in the elderly or immunocompromised patients.

Most horses also experience asymptomatic seroconversion. Only about 10 - 20 % of horses develop clinical signs after an incubation period of 3 - 15 days, of which only about 8 % are severe neurological signs. Only a few have fever as a characteristic clinical sign. Non-specific clinical signs can be: anorexia, mild fever, somnolence, colic or lameness. When meningoencephalitis develops, there are non-specific neurological signs that may be asymmetrical and progressive. Stumbling, hindlimb paralysis, head resting, dysphagia, ataxias, muscle tremors or weakness and even recumbency have been documented. Up to 40% of horses presenting with neurological signs die or have to be euthanised. Permanent damage can remain after the infection has been cured.

Diagnostic

Serology

In horses, different ELISA tests are available for the detection of IgG and IgM antibodies. IgM antibodies rise a few days post infection and remain elevated for up to 4 - 6 weeks. CAVE: IgM antibodies can be detected up to 52 days after a recent vaccination, therefore the vaccination status should be considered when assessing laboratory findings. IgG antibodies rise somewhat later and may persist longer (about 1 year). Both IgG and IgM can lead to cross-reactions with other flaviviruses (e.g. TBE virus and Usutu virus). Therefore, suspected cases must be confirmed by several virus neutralisation tests (VNT) to differentiate the flavivirus-specific antibodies of the national reference laboratory (FLI, AGES).

Direct virus detection by means of RT-PCR

Initially, there is a short viraemia, which has often already subsided by the time the

symptoms appear, so that a PCR from EDTA blood has only a very low sensitivity for diagnosis. RT-PCR from cerebrospinal fluid or brain tissue is conclusive in a positive case, but a negative PCR from cerebrospinal fluid does not exclude the infection, as the virus can also be restricted to the nerve tissue.

On post-mortem, very high levels of virus can be detected in the CNS. The use of Personal protection equipment (PPE) is important when collecting such samples. There is a case described where WNV was transmitted to a veterinary student during the dissection of a horse (removal of the brain).

Differential diagnoses

Other neurotropic infections can be considered as differential diagnoses. In particular, equine herpesvirus myeloencephalopathy due to EHV-1 or EHV-4 should be clarified in case of neurological symptoms (PCR from EDTA blood and from respiratory swabs without medium). TBE (serology IgG and IgM from serum or IgG from CSF) and Borna (PCR and/or antibodies from blood or CSF) are also possible differential diagnoses. In addition, other causes of neurological symptoms, such as injuries or poisoning, should be excluded (e.g. tetanus, botulism, listeriosis, leptospirosis).

Duty of disclosure

Infection with WNV is a zoonotic disease that must be monitored in the EU and Switzerland.

In Germany, acute infection in horses is notifiable. The basis for this is a positive IgM ELISA. Confirmation of acute infection must be carried out by the national reference laboratory due to cross-reactions of flaviviruses via VNTs. A positive IgG antibody test alone is not a basis for mandatory notification. IgG antibodies

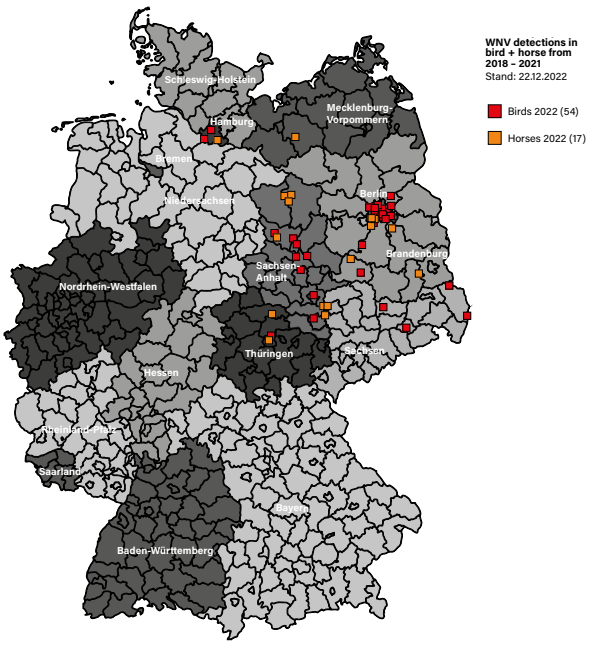

Fig. 2: Geographical distribution of reported WNV cases in horses and birds in Germany *Image source: Laboklin*

can originate from a vaccination or a longer-standing (possibly asymptomatic) infection.

Cross-reactivity must also be considered. In routine diagnostics, there is currently no ELISA test that can distinguish between antibodies against WNV from the vaccines available on the market and field virus antibodies. In Austria, notification is mandatory for all equine encephalomyelitis, regardless of its origin.

Prophylaxis

Early symptomatic treatment can significantly improve the survival rate of affected horses. Since a causal therapy is not possible, prophylactic measures come to the fore. Various vaccines are approved for the use in horses. Vaccination is recommended especially in areas where cases have already occurred in humans or animals and for horses competing abroad. The basic immunisation should be completed

Fig. 3: Mosquito larvae of Culex sp. can develop into an imago in a few days in midsummer

Image source: Dr Regina Wagner

before the start of the mosquito season. Another component of prophylaxis is to ensure protection against mosquitoes. Here, the use of repellents or protective nets/blankets should be mentioned. In addition, special attention should be paid to possible breeding sites for mosquitoes. Standing water accumulations are problematic, because in summer temperatures mosquitoes can develop from egg to larvae to imago within a few days (Figure 3). Vessels with standing water/water accumulations should be covered mosquito-proof or regularly cleaned and freshly filled (e.g. watering cans, water troughs, rainwater containers). Puddles of water, e.g. in lying car tyres or in folds of foil, should also be avoided.

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Further reading

Beck C, Jimenez-Clavero MA, Leblond A, Durand B, Nowotny N, Leparc-Goffart I, Zientara S, Jourdain E, Lecollinet S. Flaviviruses in Europe: complex circulation patterns and their consequences for the diagnosis and control of West Nile disease. Int J Environ Res Public Health. 2013 Nov 12;10(11):6049-83. doi: 10.3390/ijerph10116049.

Cavalleri JV, Korbacska-Kutasi O, Leblond A, Paillot R, Pusterla N, Steinmann E, Tomlinson J. European College of Equine Internal Medicine consensus statement on equine flaviviridae infections in Europe. J Vet Intern Med. 2022 Nov;36(6):1858-1871. doi: 10.1111/jvim.16581.

Kampen H, Holicki CM, Ziegler U, Groschup MH, Tews BA, Werner D. West Nile Virus Mosquito Vectors (Diptera: Culicidae) in Germany. Viruses. 2020 Apr 28;12(5):493. doi: 10.3390/v12050493.

Current distribution maps can be found at the Friedrich Löffler Institute (FLI):
Link: https://www.fli.de/de/aktuelles/tierseuchengeschehen/west-nil-virus/

As well as at the European Centre for Disease Control (ECDC):
Link: https://www.ecdc.europa.eu/en/west-nile-virus-infection

respectively:
https://www.ecdc.europa.eu/en/publications-data/west-nile-virus-human-cases-compared-previous-seasons-2-august-2023