



# ISWAVLD 2023

International Symposium of the World  
Association of Veterinary Laboratory  
Diagnosticians

29 JUNE-1 JULY  
2023  
Congress Centre  
Lyon

Towards  
the veterinary  
diagnostics  
of the  
future

Main topic : Surveillance and control of emerging diseases

## Improvement of molecular and serological tools for the detection of Venezuelan Encephalitis Virus (VEEV) in equids

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Venezuelan Equine Encephalitis Virus (VEEV) belongs to genus Alphavirus within family Togaviridae. The virus, transmitted by infected mosquitoes of the genera *Aedes*, *Culex* and *Psorophora*, is responsible for encephalitis in horses but also in humans. Equids serve as amplifying hosts for epizootic strains. VEEV is considered to be highly pathogenic with an equine mortality of between 40 and 90%. Historically, epizootic strains were restricted to North and West South America. However, from 1969 to 1972, an emergence occurred in Central America and the USA. Classified in categories D and E by the new animal health law, which came into force in April 2021, this disease is subject to compulsory declaration and surveillance in Europe. The European Commission is requesting systematic control of horses coming from the American continent. Under scenarios of climate change we are experiencing, and the intensification of international trade (trade, racing and international horse shows (Olympic Games 2024)), the risk of introducing VEEV into Europe and more particularly into France is not negligible. The serological diagnosis tools currently available prior to international transport or in cases of suspected Venezuelan equine encephalitis are based on time-consuming and tedious techniques that require a high level of technical expertise. In addition, they require the handling of viruses in BSL3 (Laboratory BioSafety Level 3).

As European Union Reference Laboratory (EURL) for Equine Diseases, our main objective is to develop new cost effective diagnosis tools, easy to implement, that can be deployed by all 26 Member States. For molecular diagnosis, we have optimised a sensitive and specific detection method using the VEEV/?-actin duplex RT-PCR. The specificity of the method was evaluated on different circulating strains of VEEV but also on other equine neurotropic alphaviruses and flaviviruses. The sensitivity of the method was determined. For the serodiagnosis of VEEV, we are currently developing an indirect ELISA using chimeric class 2 Sindbis virus, expressing the envelop, membrane and capsid structural proteins of VEEV on the Sindbis backbone. The performance, sensibility and sensitivity of the ELISA will be compared to that of seroneutralisation using sera from horses vaccinated or naturally infected with VEEV. The EURL will propose to the member states a range of efficient serological and molecular tools to diagnose equines infected by VEEV and therefore to alert as soon as possible public authorities in order to deploy management measures.