EXTERNAL REFERENCES

ID SCREEN® FLAVIVIRUS COMPETITION

Last update: September 2023

Publications / References:

WEST NILE VIRUS ANTIBODY DETECTION

EQUINE

1)Molini U. et al. (2022). **Neutralising antibodies to West Nile virus detected in horses in Windhoek, Namibia**. Journal of the South African Veterinary Association, 93(1), 1-3.	• 98 horse sera were investigated using the ID Screen® WNC ELISA. WNV ELISA-positive samples were subjected to a confirmatory test using a virus neutralization test (VNT). • *Results*: A total of 38 out of 98 analyzed horses tested positive using ELISA, and in seven of these, the presence of WNV-neutralizing antibodies was confirmed by VNT. The WNV seroprevalence in horses was therefore 7.14%.	Correlation with other techniques		Epidemiological study		
2)Molini U. *et al.* (2021). **West Nile Virus Seroprevalence in a Selected Donkey Population of Namibia.** Front. Vet. Sci. 8:681354.	• 260 donkey sera were tested using the ID Screen® WNC ELISA; ELISA-positive samples were confirmed using a virus neutralization test (VNT). • *Results*: A total of 108 out of 260 samples tested positive by ELISA, and in 47 out of 260 the presence of WNV-neutralizing antibodies was confirmed by VNT.	Correlation with other techniques		Epidemiological study		
3)Selim A. *et al*. (2020). **Seroprevalence and molecular characterization of West Nile Virus in Egypt.** Comparative Immunology, Microbiology and Infectious Diseases 71, 101473.	• A serological survey was conducted on 500 horse sera using the ID Screen® WNC ELISA. • *Results*: 97 out of 500 samples tested positive (seroprevalence 19.4 %).			Epidemiological study		

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4)Monaco F. *et al.* (2019). **Immunological response in horses following West Nile virus vaccination with inactivated or recombinant vaccines**. Veterinaria Italiana 55 (1), 73-79.	• To evaluate the immunological response following vaccination, WNV serologically negative horses were divided into two groups of 20 animals. One group was vaccinated (booster after 28 days) with a whole inactivated viral strain and the second group with a live recombinant canarypox virus expressing the genes coding for the WNV preM/E viral proteins. Antibodies were monitored using the ID Screen® WN IgM capture ELISA (and another commercial IgM capture Elisa), the ID Screen® WNC ELISA, and serum neutralization assay (to exclude any cross-reaction with other co-circulating related flaviviruses such as Usutu) for 360 days. • *Results*: All horses immunized with the inactivated product developed a detectable IgG response starting from the 18th-day post-vaccination (dpv) while 35 days were necessary to achieve the complete seroconversion in the group that received the recombinant vaccine IgG antibodies were detected in the animals of both groups until the end of the trial (one year after vaccination). None of the animals in the trial developed neutralizing antibodies against the Usutu virus. **The ID Screen® WNC ELISA is able to detect antibodies from the 18th-dpv for inactivated vaccine and from the 35th-dpv for recombinant vaccine until one year post-vaccination.**	Correlation with other techniques		Experimental study	Vaccination monitoring	
5)Benjelloun A. *et al.* (2017). **Seroprevalence of West Nile virus in horses in different Moroccan regions.** Veterinary Medicine and Science, 3, 198–207.	• Epidemiological study by quantifying seroprevalence of WN antibodies in horses using the ID Screen® WNC ELISA. • *Results*: Overall prevalence: 31,1%; 260/298 ELISA positive sera were confirmed in VNT.	Correlation with other techniques		Epidemiological study		
6)Haroun M. *et al.* (2017). **Occurrence of Equine West Nile Virus Among Horses in Qatar: A Preliminary Investigation.** European Scientific Journal January 2017 /SPECIAL/ edition ISSN: 1857 – 7881 (Print) e ISSN 1857-7431.	• Serological study to examine possible exposition to WNV among horses (n=260) using the ID Screen® WNC ELISA. • *Results*: seroprevalence 23,5%.			Epidemiological study		

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7)Joó K. *et al.* (2017). **Comparison of assays for the detection of West Nile virus antibodies in equine serum after natural infection or vaccination.** Veterinary Immunology and Immunopathology 183, 1–6.	• Comparison of different serological tests on naturally exposed and vaccinated horses (ID Screen® WNC ELISA, a commercial IgM capture, HIT, PRNT). • *Results*: High rate of seropositivity=16-38%; ELISA more sensitive than HIT.	Correlation with other techniques			Vaccination monitoring	Performance evaluation
8)Mehmet K. *et al*. (2017). **Serological investigation of West Nile virus infection in domestic horses and donkeys in Turkey.** Pak Vet J, 37(1): 51-54.	• Serum samples were collected from 650 domestic horses and 234 domestic donkeys and tested using the ID Screen® WNC ELISA. • *Results*: WNV seropositivity was detected at a 4.15% (27/650) level for horse serum samples and 1.28% (3/234) for donkey serum samples.			Epidemiological study		
9)Bahuon C. *et al*. (2016). **West Nile virus epizootics in the Camargue (France) in 2015 and reinforcement of surveillance and control networks.** Rev. Sci. Tech. Off. Int. Epiz., 35 (3), 811-824.	• WNV epizootics in 2015 among horses in the Camargue, using ID Screen® WNC ELISA and ID Screen® WN IgM capture ELISA, followed by confirmatory serological test VNT. • *Results*: Early detection of first equine cases, confirmed by VNT.	Correlation with other techniques		Epidemiological study		
10)Bouzalas I. *et al*. (2016). **Emergence of Equine West Nile Encephalitis in Central Macedonia, Greece, 2010.** Transboundary and Emerging Diseases. 63(6): e219-e227.	• Epidemiological study of WNV infection among horses (n=70) using the study of clinical cases, MAC-ELISA, VNT, and the ID Screen® WNC ELISA. • *Results*: Seroprevalence in asymptomatic horses = 33%.	Correlation with other techniques		Epidemiological study		
11)Chaintoutis S. *et al.* (2015). **Evaluation of cross-protection of a lineage 1 West Nile virus inactivated vaccine against natural infections from a virulent lineage 2 strain in horses, under field conditions.** Clin Vaccine Immunol 22:1040 –1049.	• Vaccination study during epidemic periods in horses, under field conditions; ID screen® WNC ELISA and/or SNT were used to check WNV in control and immunized horses. • *Results*: ID Screen® WNC ELISA confirms circulation of WNV in horse-riding clubs and efficiency of immunization. **ID Screen® WNC ELISA, a tool for monitoring WNV circulation during epidemics and the efficiency of vaccination campaigns.**	Correlation with other techniques		Epidemiological study	Vaccination monitoring	

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12)Zohaib A. *et al*. (2014). **High prevalence of West Nile virus in equines from the two provinces of Pakistan.** Epidemiol. Infect., Page 1 of 5.	• Serosurvey on WNV in an equine population (248 horses and 201 donkeys) using the ID Screen® WNC ELISA; further investigation on ELISA-positive samples was conducted using MNT (WNV, JEV, and TBEV). • *Results*: 292 sera were detected positive (seroprevalence 65%) with ID Screen® WNC ELISA; 249 sera (seroprevalence 55,4%) were found WNV positive by MNT.	Correlation with other techniques		Epidemiological study		
13)Barbić L. *et al*. (2013). **West Nile virus serosurveillance in horses in Croatia during the 2012 transmission season.** Medical Sciences, 39 (2013): 95-104.	• Active serosurveillance study of WNV in sentinel horses (n= 1472) using the ID Screen® WNC ELISA and the ID Screen® WN IgM capture ELISA. • *Results*: Acute infection was revealed with the ID Screen® WN IgM capture ELISA; seroprevalence was estimated at 8,7% with the ID Screen® WNC ELISA.			Epidemiological study		
14)Bargaoui R. *et al*. (2013). **Mapping the Serological Prevalence Rate of West Nile Fever in Equids, Tunisia**. Transboundary and Emerging Diseases, 62(1), 55–66.	• Seroprevalence study of WNV in Tunisian equids using the ID Screen® WNC ELISA and microneutralization tests (MNT). • *Results*: The overall serological prevalence rate was estimated at 28%. **The study demonstrated a perfect agreement (100%) between ID Screen® WNC ELISA and MNT.**	Correlation with other techniques		Epidemiological study		
15)Borujeni P. *et al*. (2013). **A serological survey on antibodies against West Nile virus in horses of Khuzestan province.** Iranian Journal of Veterinary Medicine IJVM 7(3):185-191.	• Evaluation of seroprevalence of WNV infection in horses using the ID Screen® WNC ELISA. • *Results*: The overall serological prevalence rate was estimated at 70,3%.			Epidemiological study		
16)Jonquiere F.*et al*. (2011). **West Nile Virus Vaccination in Horses –IgM and IgG responses after injection in different muscle.** Pferdeheilkunde 27 ,4 (July/August) 412-416.	• Study of serological response after WNV vaccination in horses with the ID Screen® West Nile Competition ELISA. • *Results*: From day 28 all samples were strongly positive and demonstrated a high humoral response to vaccination. **ID Screen® WNC ELISA, a tool to follow seroconversion, in cases of vaccination.**			Experimental study	Vaccination monitoring	

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AVIAN

17)Reemtsma H. *et al.* (2022). **Pathogenesis of West Nile Virus Lineage 2 in Domestic Geese after Experimental Infection**. Viruses, 14(6), 1319.	• 15 young geese were infected with WNV lineage 2. Seroconversion was followed using VNT and the ID Screen® West Nile Competition ELISA. • *Results*: All infected geese seroconverted at the latest 10 dpi in the ID Screen® West Nile Competition ELISA.	Correlation with other techniques			Experimental infection	
18)Talukdar A. *et al*. (2022). **Sero-prevalence of West Nile virus in urban and peri-urban poultry farms of Guwahati, India**. Frontiers in Tropical Diseases, 3, 792857.	• 864 chicken serum samples were screened using the ID Screen® West Nile Competition ELISA and further confirmed by hemagglutination inhibition (HI). • *Results*: Out of 864 samples, 36 were positive for antibodies against WNV by ELISA (4.2%) Further confirmation of these 36 ELISA-positive samples by HI revealed 27 to be positive for WNV with an overall apparent prevalence of 3.1%.	Correlation with other techniques		Epidemiological study		
19)Pallari C.T. *et al.* (2021). **Evidence of West Nile Virus seropositivity in wild birds on the island of Cyprus.** Comparative immunology, microbiology and infectious diseases, 74, 101592.	• The study investigated the seroprevalence of WNV antibodies in migratory and resident birds (836 avian blood samples comprising 44 species) using the Screen® West Nile Competition ELISA. • *Results*: Seroprevalence 1,3% (11/836 samples).			Epidemiological study		
20)Holicki C.M. *et al.* (2020). **Pathogenicity of West Nile Virus Lineage 1 to German Poultry.** Vaccines 8, 507.	• This study examined the pathogenicity of an Italian WNV lineage 1 strain after inoculation for domestic poultry (chickens, ducks, and geese). A challenge was performed on geese using WNV-infected mosquito bites. Sera were analyzed using VNT. For confirmation, the samples were also screened using the ID Screen® WNC ELISA. • *Results*: Seroconversion was observed in all three poultry species after inoculation (from 10 dpi), measured via VNT and the ID Screen® WNC ELISA. Four of the eight geese exposed to the WNV-positive mosquitoes became infected and seroconverted. **ID Screen® WNC ELISA, a tool to follow seroconversion, in cases of vaccination, as well as VNT.**	Correlation with other techniques			Experimental infection	

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21)Islam A. *et al.* (2020). **Serological Evidence of West Nile Virus in Wild Birds in Bangladesh.** Vet. Sci. 2020, 7, 164.	• 184 sera samples from wild resident and migratory birds (19 identified species) were tested using the ID Screen® WNC ELISA. • *Results*: seroprevalence 11.9% (22 sera tested positive).			Epidemiological study		
22) Ain-Najwaa M.Y. *et al.* (2020). **Evidence of West Nile virus infection in migratory and resident wild birds in west coast of peninsular Malaysia.** One Health 10, 100134.	• Sera for 155 migratory birds were tested using the ID Screen® WNC ELISA. Cross-reactivity towards Japanese Encephalitis virus (JEV) was also carried out using a JEV-double antigen sandwich (DAS) ELISA. • *Results:* Of 155 serum samples analyzed using the ID Screen® WNC ELISA, 30 were positive. Out of these serum samples, only one sample showed a positive reaction using the JEV DAS-ELISA. Therefore, this study found a positive reaction for anti-WNV IgG antibodies in 18.71% (29/155).			Epidemiological study		
23) Kim C-Y. *et al.* (2016). **First detection of West Nile virus in domestic pigeon in Korea.** J Vet Sci, 17(4), 587-589.	• 75 sera samples from pigeons were tested using the ID Screen® WNC ELISA. • *Results*: out of 75 samples, 3 pigeons tested positive. WNV presence. was confirmed with nested reverse transcription polymerase chain reaction analysis and sequencing.	Correlation with other techniques		Epidemiological study		
24)Pastiu A. *et al.* (2016). **Wild Birds in Romania Are More Exposed to West Nile Virus Than to Newcastle Disease Virus.** Vector-Borne and Zoonotic Diseases, 16(3), 176-180.	• Evaluation of seroprevalence of WNV in wild birds (n=159) and domestic birds (n=21) from Romania with ID Screen® WNC ELISA. • *Results*: Seroprevalence in wild birds= 32,14% and seroprevalence in domestic birds = 19,05%.			Epidemiological study		
25)Hammouda A. *et al.* (2015). **Exposure of resident sparrows to West Nile virus evidenced in South Tunisia.** Epidemiol. Infect., 143, 3546–3549.	• Investigation of exposure of wild sparrows (n=208) through the ID Screen® WNC ELISA and VNT (to discriminate WNV and USUV). • *Results*: Seroprevalence = 1%, sera found positive with ID Screen® WNC ELISA were confirmed WN specific with VNT.	Correlation with other techniques		Epidemiological study		

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26)Chaintoutis S. *et al*. (2014). **Evaluation of a West Nile virus surveillance and early warning system in Greece, based on domestic pigeons.** Comparative Immunology, Microbiology and Infectious Diseases 37, 131– 141.	• After a WNV epidemic, a surveillance system based on serological testing of domestic pigeons with ID Screen® WNC ELISA; samples positive by ELISA were further assayed by VNT against USUV. Study 1: n=655, study 2: n=210. • *Results*: Study 1: seroprevalence 38%; study 2: seroprevalence 31%. All samples assayed by VNT were WNV confirmed except one, USUV confirmed.	Correlation with other techniques		Epidemiological study		
27)Chaskopoulou A. *et al.* (2013). **Detection and Early Warning of West Nile Virus Circulation in Central Macedonia, Greece, Using Sentinel Chickens and Mosquitoes.** Vector Borne Zoonotic Dis. 13(10):723-32.	• WNV seroprevalence study in sentinel chickens with ID Screen® WNC ELISA; ELISA-positive sera were further tested using micro-PRNT. • *Results*: 11/47 chicken seroconverted; all positive samples were confirmed by micro-PRNT.	Correlation with other techniques		Epidemiological study		
28)Ziegler *et al*. (2013). **Pathogenesis of West Nile virus lineage 1 and 2 in experimentally infected large falcons.** Veterinary microbiology, 161(3-4), 263-273.	• To investigate the WNV pathogenesis, twelve large falcons were inoculated with viruses belonging to two different lineages (lineage 1 strain NY 99 and lineage 2 strain Austria). Serology was followed using the ID Screen® WNC ELISA. • *Results*: Seroconversion of experimentally infected falcons was detected at 6 dpi with the ID Screen® WNC ELISA.				Experimental study	
29) Sotelo E. *et al*. (2011). **Pathogenicity of two recent Western Mediterranean West Nile virus isolates in a wild bird species indigenous to Southern Europe: the red-legged partridge.** Veterinary Research 42:11.	• Red-legged partridges were experimentally infected with two WNV isolates; ID Screen® WNC ELISA and RT-PCR were used prior to the experiment to ensure that exposure to WNV had not occurred; ELISA and VNT were then used to follow antibody response. • *Results*: In inoculated partridges, specific antibodies to WNV as revealed by ID Screen® WNC ELISA were observed slightly earlier (6dpi) than neutralizing antibodies (VNT).	Correlation with other techniques			Vaccinationn monotoring	

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AVIAN AND HORSES

30)Raleigh P. *et al*. (2012). **Surveillance for antibodies to West Nile virus in Ireland.** Veterinary Record 170: 180.	• Serological study of WNV in birds and horses (from field or vaccination experiments) using the ID Screen® WNC ELISA, a commercial ELISA, MVNT and PRNT. • *Results*: All the post-vaccination sera were positive (up to 149 dpi in chicken); good agreement between the ELISAs; MVNT and PRNT confirmed results**.**	Correlation with other techniques			Experimental study	Performance evaluation

OTHER SPECIES

31)Molini U. *et al*. (2023). **Low Seroprevalence of WNV in Namibian Dogs Suggests a Limited Effectiveness as Sentinels for Infection Monitoring**. Tropical Medicine and Infectious Disease, 8(4), 203.	• 426 domestic **dog** samples were analyzed using the ID Screen® WNC ELISA and VNT. • *Results*: A total of 70/426 (16.43%) analyzed dog samples tested positive via ELISA, and the presence of WNV neutralizing antibodies was confirmed via VNT in 12 animals.	Correlation with other techniques		Epidemiological study		
32)Cosseddu G.M. *et al*. (2021). **Sero-surveillance of emerging viral diseases in camels and cattle in Nouakchott, Mauritania: an abattoir study.** Tropical Animal Health and Production, 53(2), 1-6.	• 159 **camel** sera were tested using the ID Screen® WNC ELISA. Samples, which resulted positive were tested with the confirmatory virus neutralization test (VNT). • *Results*: Seroprevalence 92% (146 sera out 159) prevalence of WNV; 41 camel sera out of 146 were confirmed to be WNV positive by VNT.	Correlation with other techniques		Epidemiological study		
33)Mohammed M.N. *et al*. (2021). **Serological evidence of West Nile viral infection in archived swine serum samples from Peninsular Malaysia**. J Vet Sci.22(3):e29.	• 80 archived **swine** serum samples were tested using the ID Screen® WNC ELISA; a DAS ELISA kit – Porcine JE-IgG ELISA kit was used to screen samples against the endemic cross-reacting JEV. • *Results*: Seroprevalence 62.5% (50/80 sera) Out of the 50 positive sera, only one sample was positive for Japanese encephalitis virus antibodies.			Epidemiological study		

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34)Pham-Thanh L. *et al.* (2021). **Dogs as Sentinels for Flavivirus Exposure in Urban, Peri-Urban and Rural Hanoi, Vietnam.** Viruses 13, 507.	• Cross-sectional study to estimate seroprevalence of flavivirus among **dogs** (n=475). • *Results:* The overall flavivirus seroprevalence in the dog population was 70.7%.			Epidemiological study		
35)Ain-Najwa M.Y. *et al.* (2020). **Exposure to Zoonotic West Nile Virus in Long-Tailed Macaques and Bats in Peninsular Malaysia.** Animals 2020, 10, 2367.	• Sera from 81 **long-tail macaques** were tested using the ID Screen® WNC ELISA. The samples, which resulted positive were subjected to JEV screening using a specific double-antibody sandwich ELISA to rule out cross-reactivity. • *Results*: 24 of the 81 sera tested were positive (seropositivity: 29.63%); none of the samples showed a positive reaction towards JEV.			Epidemiological study		
36)Dinç E. *et al.* (2020) **Serological Investigation of West Nile Virus (WNV) Infection in Cats and Dogs.** ARRB, 35(1): 65-71.	• Sera from 82 **cats** and **246** dogs were tested using the ID Screen® WNC ELISA. • *Results*: presence of antibodies was detected in 0.41% of the tested dog blood sera (1/246) and in 1.22% of the cat blood sera (1/82).			Epidemiological study		
37)Selim A. *et al.* (2020). **The first detection of anti-West Nile virus antibody in domestic ruminants in Egypt.** Tropical Animal Health and Production, 52(6), 3147-3151.	• 360 serum samples (100 **cattle**, 50 **buffalo**, 50 **camels**, 85 **sheep**, and 75 **goats**) were examined using the ID Screen® WNC ELISA. • *Results*: Seroprevalence 22%, 0%, 40%, 3.5%, and 5.3% in cattle, buffalo, camel, sheep, and goats, respectively.			Epidemiological study		
38)Montagnaro S. *et al.* (2019). **Serological evidence of mosquito-borne flaviviruses circulation in hunting dogs in Campania Region, Italy**. Vector-Borne and Zoonotic Diseases, 19(2), 142-147.	• 183 hunting **dog** sera were analyzed using the ID Screen® WNC ELISA. Positive ELISA samples were further analyzed by SNT using the USUTU virus and West Nile virus. • *Results*: overall seroprevalence value of 40.43% (74/183), using the ID Screen® WNC ELISA. Among the 74 ELISA-positive sera, SNT showed that 24 sera resulted positive for Usutu virus with an overall prevalence of 13.11% (24/183) but none of the ELISA-positive samples resulted positive for West Nile virus.	Correlation with other techniques		Epidemiological study		
39)Vitaskova E. *et al.* (2019). **Serologic Survey of Selected Viral Pathogens in Free-Ranging Eurasian Brown Bears (*Ursus arctos arctos*) from Slovakia.** Journal of wildlife diseases, 55(2), 499-503.	• Sera of 24 **European brown bears** were tested using the ID Screen® WNC ELISA. Positive samples were subsequently confirmed by virus neutralization test for WNV and TBEV. • *Results*: 1/24 of the bears was positive (seropositivity 4%). The positive sample was further examined by VNT and was found to be positive for WNV but negative for TBEV.			Epidemiological study		

40)Hassine T.B. *et al*. (2017). **Emerging vector-borne diseases in dromedaries in Tunisia: West Nile, Blue Tongue, Epizootic Haemorrhagic disease and Rift Valley fever.** Onderstepoort Journal of Veterinary Research 84(1), a1316.	• 118 sera from **dromedaries** were tested with ID Screen® WNC ELISA. • *Results*: Seroprevalence=25,8%.			Epidemiological study		
41)Pâslaru A. *et al*. (2016). **West Nile Virus Serosurveillance in Wild Boars from the East of Romania.** Bull. UASVM Vet. Med, 73, 144-148.	• Assessment of WNV circulation in **wild boars** (n=68) using the ID Screen® WNC ELISA. • *Results*: seroprevalence in the two regions respectively 53,33% and 71,05%.			Epidemiological study		
42)Lan D. *et al*. (2011). **Serological evidence of West Nile virus in dogs and cats in China.** Arch Virol 156:893-895.	• WNV seroprevalence study in **dogs** (n=367) and **cats** (n=309) using the ID Screen® WNC ELISA. • *Results*: Dogs: seroprevalence 5,7%; when ELISA positive samples were tested by PRNT, 4,6% samples were positive and two samples were JEV positive; seroprevalence cats: 15,9%; among ELISA positive samples, 14,9% were tested positive by PRNT.			Epidemiological study		

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HUMANS

43)Tinto B. *et al*. (2022). **Screening of circulation of Usutu and West Nile Viruses: A one health approach in humans, domestic animals and mosquitoes in Burkina Faso, West Africa**. Microorganisms, 10(10), 2016.	• **500 human sera samples from blood donors** and 204 sera samples from animals (81 **horses**, 52 **dogs**, 50 **chickens**, and 21 **pigeons**) were studied using the ID Screen® WNC ELISA and microneutralization tests (MNT) against WNV and USUV. • *Results*: **Human samples: A total of 389 samples were positive (77.65%); samples positive for ELISA were then tested using MNT against WNV and USUV. Among the positive samples, 71 tested positive for USUV-specific antibodies (14.17%), and 96 for WNV-specific antibodies (19.16%)., 95% CI: 15.95–22.83). A total of 52 samples demonstrated neutralizing activity for both USUV and WNV**. Domestic animals: Among the 81 horses analyzed, 73 horses tested positive for ELISA. Among them, 5 (6.17%), were positive for USUV antibodies, and 14 (17.28%) for WNV antibodies. Anti-flavivirus antibodies were identified in 14 dogs; only one of the dogs presented neutralizing activity for WNV; and all dog samples were negative for USUV antibodies. Concerning avian analyses, anti-flavivirus antibodies were identified in 4 chicken samples but no positives for USUV and WNV after MNT tests. In pigeons, of the two samples that gave a positive result for the ELISA test, one sample presented a USUV-positive antibody and another one presented anti-WNV antibodies. **The ID Screen® WNC ELISA is able to detect flavivirus (WNV and USUV) antibodies in humans.**	Correlation with other techniques	Particular species	Epidemiological study		
44)Constant O. *et al*. (2022). **One Health surveillance of West Nile and Usutu viruses: A repeated cross-sectional study exploring seroprevalence and endemicity in Southern France, 2016 to 2020.** Eurosurveillance, 27(25), 2200068.	• Prevalence study of WNV and USUV in a repeated cross-sectional study by serological and molecular analyses of **human**, **dog**, **horse**, bird, and mosquito samples Serological study on dogs (n=149), horses (n=135), and human (n=500) sera was performed using the ID Screen® WNC ELISA. To confirm **WNV or USUV** infection, viral microneutralization tests (MNT) were performed on ELISA-positive samples. • *Results*: **Human sera: 53 of the 500 samples were identified positive using the ID Screen® WNC ELISA. Among these samples, 15 tested positive for USUV-specific antibodies (3%; 95% CI: 1.5–4.5) and six for WNV-specific antibodies (1.2%; 95% CI: 0.25–2.15). One sample demonstrated neutralizing activity for both WNV and USUV. Dog sera: presence of flavivirus antibodies using the ID Screen® WNC ELISA was**	Correlation with other techniques	Particular species	Epidemiological study		

	detected in three dog samples analyzed (1.63%; 95% CI: 0.00–3.46): two USUV antibody-positive samples were detected by MNT (1.08%; 95% CI: 0.00–2.58). One dog was positive for WNV antibodies (0.54%; 95% CI: 0.00–1.60). Horse sera: 40 out of 235 sera identified positive using the ID Screen® WNC ELISA (17.02%; 95% CI: 12.21– 21.82). **Among these samples, nine were positive for USUV-specific antibodies (3.83%; 95% CI: 1.37–6.28) and 31 for WNV specific antibodies (13.19%; 95% CI: 8.86–17.51).** **ID Screen® WNC ELISA is able to detect flavivirus (WNV and USUV) antibodies in humans.**					
45)Niczyporuk J. *et al*. (2015). **Occurrence of West Nile Virus Antibodies in Wild Birds, Horses, and Humans in Poland.** BioMed Research International vol 2015, Article ID 234181.	• Study on detection of WNV antibodies in serum samples from **wild birds** (n=474), **horses** (378) and **humans** with neurological symptoms (n=42) by ID Screen® WNC ELISA, ID Screen® WN IgM capture ELISA, and another commercial blocking ELISA; positive results in bird samples were submitted to VNT. • *Results*: The study revealed WNV antibodies in 63 samples from birds (with ID Screen® WNC ELISA and another commercial test), 1 from horses, and 14 from human samples; all positive samples were negative with ID Screen® WN IgM capture ELISA but were confirmed positive with VNT. **ID Screen® WNC ELISA is able to detect WNV antibodies in humans.**	Correlation with other techniques	Particular species	Epidemiological study		

FLAVIVIRUS ANTIBODY DETECTION

PANFLAVIVIRUS WNV/TBEV/USUV/JEV

46)Gothe L.M.R. *et al.* (2023). **Horses as Sentinels for the Circulation of Flaviviruses in Eastern–Central Germany**. Viruses 2023, 15, 1108.	• sera from 1232 unvaccinated **horses** were tested using ID Screen® WNC ELISA. To estimate the true seropositive ratio of infection with WNV, TBEV, and USUV, positive and equivocal results were confirmed by a virus neutralization test (VNT) • *Results*: In total, 125 horse sera reacted positive in ID Screen® WNC ELISA. Based on the VNT, 40 sera showed neutralizing antibodies against **WNV**, 69 against **TBEV**, and 5 against **USUV**. 3 sera showed antibodies against more than one virus, and 8 were negative based on the VNT. The overall seropositive ratio was 3.3% for WNV, 5.6 for TBEV, and 0.4% for USUV infections.			Epidemiological study		
47)Bergmann F. *et al.* (2022). **Seroepidemiological Survey of West Nile Virus Infections in Horses from Berlin/Brandenburg and North Rhine-Westphalia, Germany**. Viruses, 14(2), 243.	• 437 **equine sera** were screened for WNV-specific antibodies using the ID Screen® WNC ELISA and virus/specific neutralization tests (VNT) were performed on ELISA-positive and doubtful samples. • *Results*: 52 sera were ELISA positive; 35 sera were confirmed by **WNV**-VNT, 6 sera were confirmed by **TBEV**-VNT, and 1 serum was confirmed by **USUV**-VNT.	Correlation with other techniques		Epidemiological study		—
48)Ganzenberg S. *et al.* (2022). **Seroprevalence and risk factors for equine West Nile Virus infections in eastern Germany, 2020**. Viruses, 14(6), 1191.	• 940 **horse sera** were tested using the ID Screen® WNC ELISA and reactive samples were further tested by the ID Screen® WNIGM capture ELISA and confirmed by virus neutralization test (VNT). • *Results*: 106 serum samples showed antibodies against flaviviruses using the ID Screen® WNC ELISA, of which six tested positive for WNV IgM. The VNT verified a **WNV** infection for 54 samples (50.9%), while 35 sera neutralized **TBEV** (33.0%), and 8 sera neutralized **Usutu** virus (7.5%).	Correlation with other techniques		Epidemiological study		—

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49)Beck C. *et al*. (2017). **Improved reliability of serological tools for the diagnosis of West Nile fever in horses within Europe**. PLoS Negl Trop Dis 11(9): e0005936.	• Inter-laboratory proficiency tests were held in 2010 and 2013 to evaluate WNV serological diagnostic tools suitable for European national reference laboratories. • *Results*: ID Screen® WNC ELISA: Specificity 100%, high analytical sensitivity =100% for flaviviruses (**WNV**, **USUTU**, **JEV**, **TBEV**).	Correlation with other techniques				Performance evaluation

PANFLAVIVIRUS WNV/USUV/BAGAZA VIRUS

50) Jurisic L. *et al*. (2023). **Immunization with Usutu virus and with a chimeric West Nile virus (WNV) harboring Usutu-E protein protects immunocompetent adult mice against lethal challenges with different WNV lineage 1 and 2 strains**. Veterinary Microbiology, 277, 109636.	• Experimental infection on **mice** with Usutu virus and with a chimeric West Nile virus (WNV) harboring Usutu-E protein; serological screening using the ID Screen® WNC ELISA and SN for neutralizing USUV and WNV antibodies was performed on serum samples collected. • *Results*: mice primed with high dose USUV were either positive (93%) or doubtful (7%) at 21 dpi when using the ID Screen® WNC ELISA. Mice primed with low dose **USUV** showed lower level of ELISA positivity as only the 20% was positive, 20% doubtful and the remaining 60% was negative. Samples of mice primed with USUV and challenged with WNV did not show, apparently, USUV antibodies at 21 days post challenge when USUV was used as antigen in the SN assay. Mice primed with **WNV**E-USUV started to succumb by 7 dpi and 82% survival was observed 30 out of 37 mice showed neutralizing antibodies against USUV at 21 dpi before challenge with a GMT of 15. These mice showed a widespread ELISA-positivity as 95% was positive and 5% of mice were doubtful.	Correlation with other techniques			Experimental infection	
51)Folly A. *et al*. (2022). **Evidence for overwintering and autochthonous transmission of Usutu virus to wild birds following its redetection in the United Kingdom**. Transboundary and Emerging Diseases, 69:3684–3692.	• 86 serum samples taken from **captive birds** were tested using the ID Screen® WNC ELISA; positive ELISA samples were also subjected to PRNT using USUV. • *Results*: Of the 86 serum samples, 15 tested positive for flavivirus antibodies using a competition ELISA. Of these ELISA-positive samples, 12 were screened by PRNT, and 10 serum samples from 7 birds (included pelicans, griffon vultures, Humboldt penguins, pigeons) had **USUV**-specific neutralizing antibodies.	Correlation with other techniques	Particular species	Epidemiological study		

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52) Vasic A. *et al*. (2022). **West Nile virus in the Republic of Serbia—Diagnostic performance of five serological tests in dog and horse sera.** Transbound Emerg Dis. 1–10.	• Seroprevalence study for WNV in clinically healthy **dog** (n=184) and **horse** sera (n=232) comparing diagnostic values of 'in-house' and commercially available indirect immunofluorescence (IFA) and the ID Screen® WNC ELISA tests to 'gold standard' virus neutralization test (VNT). Sera were further tested for Usutu virus and tick-borne encephalitis virus in VNT (**WNV and USUTUV**). • *Results*: Comparative results of diagnostic tests in dogs ranged from 18.7% seropositivity by an 'in-house' ELISA to 31.9% by commercially available ELISA. In horses, seropositivity ranged from 36.2% by 'in-house' IFA to 32.5% by a commercially available IFA and from 26.3% by 'in-house' IgG ELISA to **20.9% by the ID Screen® WNC ELISA. Seropositivity was confirmed by VNT in 36.9 % of tested dog sera while in four dog and seven horse sera, positivity resulted from Usutu virus infection**.	**Correlation with other techniques**		**Epidemiological study**		**Performance evaluation**
53)Napp S. *et al.* (2021). **Widespread circulation of flaviviruses in horses and birds in Northeastern Spain (Catalonia) between 2010 and 2019**. Viruses, 13(12), 2404.	• Serum samples from **birds** (n=3791) and **horses** (n= 1856) were analyzed using the ID Screen® WNC ELISA; positive samples were further tested by microneutralization test (MNT) against **WNV**, **USUV**, **Bagaza virus** (for birds), and TBEV (for horses). • *Results*: 10.0% (380/3791) of the samples from birds and 9.8% (182/1856) of the samples from horses tested positive for ELISA. Of the 205 serum samples from birds tested by MNT, 118 showed specific antibodies for WNV, 19 for USUV, and 68 were classified as undetermined flavivirus. None were classified as BAGV infection. Of the 164 sera from horses tested by MNT, 92 were positive for WNV, 11 for USUV, 4r for TBEV, and 57 were classified as infected by an undetermined flavivirus.	**Correlation with other techniques**		**Epidemiological study**		

54)Assaid N. *et al.* (2020). **Evidence of circulation of West Nile virus in *Culex pipiens* mosquitoes and horses in Morocco.** Acta Tropica 205, 105414.	• 92 **horse** sera were tested using the ID Screen® WNC ELISA. Flavivirus microsphere immunoassay (MIA, a test which allows distinguishing between the three flaviviruses: WNV, USUV, and TBEV) was performed on ELISA-positive samples; flaviviruses identified by MIA and all undetermined ELISA-positive flavivirus samples were investigated using MNT against WNV and USUV. • *Results*: The seroprevalence by the ID Screen® WNC ELISA was 33.7% (31/92). The MIA test showed that 25.0% (23/92) were positive for **WNV**, 2.1% (2/92) were positive for **USUV**, 4.3% (4/92) were positive for undetermined flavivirus and 2.1% (2/92) were positive for both WNV and USUV. The MNT showed that 30.4% (28/92) were positive for WNV; samples positive for USUV, 2 positives for both WNV and USUV, and one for undetermined flavivirus by MIA were found WNV positive by MNT. All sera were negative for USUV in MNT. **The ID Screen® WNC ELISA is well correlated with MIA and MNT do detect antibodies against flaviviruses.**	Correlation with other techniques		Epidemiological study		
55) Constant O. *et al*. (2020). **Evidence of Exposure to USUV and WNV in Zoo Animals in France.** Pathogens 9(12), 1005.	• 411 sera from 70 species (**captive birds**, n=137 and **mammals**, n=274) were tested using the ID Screen® WNC ELISA. The positive sera were then tested using virus-specific microneutralization tests against **USUV and WNV**. When positive samples of ELISA were found negative in WNV and USUV MNTs, **TBEV and MEAV virus** neutralization assays were carried out. • *Results*: **antibodies against flaviviruses were identified in 23 out of 137 birds (seropositivity 16.79%). Among them, 20 specimens tested positive for USUV-specific antibodies by MNT (seropositivity 14.59%) and 2 specimens for WNV-specific antibodies (seropositivity1.45%). A total of 11 mammals were positive with the ELISA kit (seropositivity 4.01%). Most of the positive mammals had antibodies against USUV, including one Asian lion, one maned wolf, one Iberian wolf and two African wild dogs. Antibodies against WNV were found in one *dama* gazelle and 3 animals presented antibodies against non-identified flaviviruses** (the corresponding sera turned out to be negative in TBEV and MEAV neutralization assays).	Correlation with other techniques	Test of particular species	Epidemiological study		

56) Llorente F. *et al*. (2019). **Influence of flavivirus co-circulation in serological diagnostics and surveillance: A model of study using West Nile, Usutu and Bagaza viruses.** Transboundary and emerging diseases, 66(5), 2100-2106.	• **Red-legged partridges** were experimentally infected with **WNV (lineages 1 and 2), USUV or BAGV** and further aznalyzed using the ID Screen® WNC ELISA and another avalaible commercial WN competition ELISA, and by the confirmatory virus neutralization test (VNT). • *Results*: The two ELISA kits examined reacted positively with all serum samples from WNV-infected partridges However, differences arose when analysing sera obtained from birds inoculated with heterologous flaviviruses (USUV and BAGV): while the other commercial kit gave negative results in all sera from USUV-infected partridges (n = 8) and just one positive reaction and two doubtful results out of 12 sera from BAGV-infected partridges**, the IDvet kit reacted with all non-WNV flavivirus sera (seven from USUV-infected partridges and five from BAGV-infected partridges).** • **The ID Screen® WNC ELISA is better than the other available commercial WN Competition ELISA for detecting a wide spectrum of flaviviruses.**	Correlation with other techniques		Epidemiological study		
57)Montagnaro S. *et al*. (2019). **Serological evidence of mosquito-borne flaviviruses circulation in hunting dogs in Campania Region, Italy.** Vector-Borne and Zoonotic Diseases, 19(2), 142-147.	• A flavivirus survey was performed on 183 **hunting dogs** using the ID screen® WNC ELISA. To identify the circulating Flavivirus, **WNV and USUV** seroneutralization assays were performed on ELISApositive samples. • *Results:* Seroprevalence 40.43% (74/183). Among the 74 ELISA-positive sera, seroneutralization test showed that 24 sera resulted positive for Usutu virus with an overall prevalence of 13.11% (24/183) but none of ELISA-positive samples resulted positive for West Nile virus.	Correlation with other techniques	Test of particular species	Epidemiological study		
58)Napp S. *et al*. (2019). **Usefulness of Eurasian Magpies (*Pica pica*) for West Nile virus Surveillance in Non-Endemic and Endemic Situations.** Viruses 11, 716.	• A multiyear series of cross-sectional surveys for WNV were carried out in **Eurasian magpies** using the ID screen® WNC ELISA. Positive samples were screened for confirmation by SNT for **WNV and Bagaza virus (BAGV).** • *Results*: August 2017: **out of 9 magpies tested, 2 were positive (seroprevalence 22%), these 2 cases were confirmed WNV positive by SNT.** 2018-2019: **seroprevalence flaviviruses 44-83%, seroprevalence WNV 8-44%, ; one of the magpies sampled positive by ELISA was tested positive for Bagaza virus (BAGV) by SNT**.	Correlation with other techniques		Epidemiological study		

59)Lim S.M. *et al*. (2018). **Serologic evidence of West Nile virus and Usutu virus infections in Eurasian Coots in the Netherlands.** Zoonoses and public health, 65(1), 96-102.	• 265 serum samples from **wild birds** were screned using the ID screen® WNC ELISA. Elisa positive samples were further tested using VNT (against **WNV-Ita09, WNV-Greece- 1 or USUV-939/01** strains). • *Results*: **Of the 265 screened serum samples, 27 were found to be WNV–antibody-positive using the ID screen® WNC ELISA and subsequent cross-neutralization experiments using WNV and USUV confirmed that five serum samples were positive for only WNV-neutralizing antibodies and seven for only USUV**.	Correlation with other techniques		Epidemiological study		

TICK-BORNE ENCEPHALITIS VIRUS (TBEV)

60)Gonzalez G. *et al*. (2022). **A One-Health Approach to Investigating an Outbreak of Alimentary Tick-Borne Encephalitis in a Non-endemic Area in France (Ain, Eastern France): A Longitudinal Serological Study in Livestock, Detection in Ticks, and the First Tick-Borne Encephalitis Virus Isolation and Molecular Characterisation.** Frontiers in microbiology, 13, 863725-863725.	• One health study performed during one year about an outbreak of tick-born encephalitis (TBE) in France. Serology was followed in suspected animals (**goats** and **cows**) using the ID Screen® WNC ELISA (interpretation was modified for sera close to the doubtful and negative thresholds, with an extension of the doubtful interval, to ascertain the detection of sera with low-TBEV antibody levels by ELISA and then increase sensitivity). Samples with positive and doubtful results in ELISA were then tested for the presence of specific neutralizing antibodies against **TBEV** by micro virus neutralization tests. An in-house IgM-capture enzyme immunoassay (MAC-ELISA) with whole inactivated TBEV was also performed. • *Results*: IgG and IgM antibodies against TBEV were detected using the ELISA tests (ID Screen® WNC ELISA and IgM capture) and MNT.	Correlation with other techniques		Epidemiological study		

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61)Bournez L.*et al.* (2020). **Exposure of Wild Ungulates to the Usutu and Tick-Borne Encephalitis Viruses in France in 2009–2014: Evidence of Undetected Flavivirus Circulation a Decade Ago**. Viruses 12(1), 10.	• Serum samples from **wild boar** and **roe deer** were screened with ID Screen® WNC ELISA. Samples found positive or doubtful were then tested for antibodies against **WNV, BAGV, USUV and TBEV/LIV** by MIA and MNT. • *Results*: **Panflavivirus antibodies: 5,6% in wild boars and 2,1% in roe deer; Usutu and TBEV/LIV antibodies were then detected in both species**. ELISA positive or doubtful results could not be confirmed by MNT or MIA due to serum quality or the lower analytical sensitivity of these tests, which is consistent with the low antibody titers (high % S/N) found by cELISA for most of these serum samples. **ID Screen® WNC ELISA, used as a panflavivirus cElisa to screen samples before specific flavivirus detection**.	Correlation with other techniques		Epidemiological study		
62)Beck C. *et al.* (2015). **A High-Performance Multiplex Immunoassay for Serodiagnosis of Flavivirus-Associated Neurological Diseases in Horses.** BioMed Research International Volume 2015, Article ID 678084.	• Description of a multiplex immunoassay compared to MNT, PRNT and ID screen® WNC ELISA to detect WNV antibodies in **horses**. • *Results*: **ID Screen® WNC ELISA recognizes all the WN, JEV and TBEV infected sera.** **ID Screen® WNC ELISA, a tool to detect 3 flaviviruses: WNV, JEV and TBEV.**	Correlation with other techniques				
63)Rushton J. *et al.* (2013). **Tick-borne Encephalitis Virus in Horses, Austria, 2011.** Emerging Infectious Diseases Vol. 19, No 4.	• Study to determine status of TBEV infection in **horses** in Austria, using ID Screen® WNC ELISA for screening antibodies against flaviviruses; ELISA-positive samples were further investigated by using virus-specific neutralization assays for **WNV, USUV, and TBEV**. • *Results*: **26,1% of horses were positive with ID Screen® WNC ELISA, all these samples were positive for TBEV by virus-specific neutralization test.**	Correlation with other techniques		Epidemiological study		

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JAPANESE ENCEPHALITIS VIRUS (JEV)

64)Ruget A. S. *et al.* (2018). **Japanese encephalitis circulation pattern in swine of northern Vietnam and consequences for swine's vaccination recommendations.** Transboundary and emerging diseases. 65(6), 1485-1492.	• 641 **pigs** sera were analyzed for Japanese encephalitis virus (**JEV**) using the ID screen® WNC ELISA. A subset of ELISA-positive samples was confirmed by the JEV neutralization test compared with the West Nile virus neutralization test. • *Results*: **Three hundred and eighty-seven (60.4%) and 28 (4.4%) samples of 641 tested positive and doubtful by ELISA, respectively. A set of 108 flavivirus ELISA-positive sera were tested in JEV and WNV MNTs. JEV infection was evidenced in 105 pigs (97.2%)**	Correlation with other techniques Test of particular species Epidemiological study

ZIKA AND DENGUE VIRUSES

65)Tinto B. *et al*. (2022). **Serological Evidence of Zika Virus Circulation in Burkina Faso.** Pathogens 11, 741.	• A total of 501 **human sera** were first tested using the ID Screen® WNC ELISA to detect flavivirus antibodies. Positive sera were then tested using Luminex to detect **ZIKV** and **the four serotypes of DENV** antibodies and virus-specific microneutralization tests against ZIKV were performed. • *Results*: From those 501 serum samples screened by ELISA to detect prior flavivirus infection, antibodies against flaviviruses were identified in 400 samples (79.84%). These ELISA-positive samples were then tested with Luminex: 229 samples were found positive for ZIKV (representing 45.70% of the total samples) 98 positives for DENV-1 (19.56%), 280 positives for DENV-2 (48.86%), 204 DENV-3 positive (40.71%) and 199 DENV-4 positive (39.72%). All ZIKV Luminex-positive samples were tested by microneutralization tests: 114 samples were found positive (22.75%). **ID Screen® WNC ELISA able to detect flavivirus (ZIKV and the four serotypes of DENV) antibodies in humans.**	Correlation with other techniques Epidemiological study

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66)Beck C. *et al*. (2019). **Serological evidence of infection with dengue and Zika viruses in horses on French Pacific Islands.** PLoS Negl Trop Dis 13(2): e0007162.	• **Horse** sera were tested to highlight flaviviruses: sera were first identified with ID Screen® WNC ELISA then confirmed with MIA, then tested with MNT (**DENV, ZIKV, WN, JEV**). • *Results*: Identification of DENV and ZIKV sera. **ID Screen® WNC ELISA, a tool to detect ZIKAV and DENV in horses in endemic areas.**	Correlation with other techniques		Epidemiological study		
67)Dolz G. *et al.* (2019). **Detection of antibodies against flavivirus over time in wild non-human primates from the lowlands of Costa Rica.** PLoS One, 14(7), e0219271.	• A total of 209 sera from free-range **non-human primates** were tested using the ID screen® WNC ELISA. Sera sampled in 2010-2012 tested positive were tested in PCR for **DENV** and **WNV**. • *Results*: **A total of 53 (25.4%) out of 209 individuals yielded positive results in ELISA,** while only two animals (1.0%) showed inconclusive reactions**. A total of four (5.6%) out of 71 blood samples yielded positive results in PCR to DENV2 (one animal), DENV3 (one animal) and DENV4 (two animals). Furthermore, three samples (4.2%) yielded positive results in RT-PCR to WNV**.	Correlation with other techniques		Epidemiological study		

TEMBUSU VIRUS

68)Lin J. *et al*. (2015). **Efficacy evaluation of an inactivated duck Tembusu virus vaccine**. Avian diseases, 59(2), 244-248.	• Evaluation of the potential use of an inactivated virus-based vaccine for the control and prevention of the newly emerged duck **Tembusu virus** infection in **ducks and geese**. Seroconversion was followed using the ID Screen® WNC ELISA. • *Results*: the study showed that more than 80% of immunized ducks were protected against virulent virus challenge. The protection is also correlated with a positive virus-specific antibody response as detected by ELISA. In contrast, none of the control ducks and geese had any detectable antibody response.		Test of particular species		Experimental study	

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