

Seroprevalence of West Nile Virus in Equine and Chickens in Sudan

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Abstract

West Nile virus (WNV) mainly infects birds, horses and human. 736 serum samples from horses, donkeys and chickens from Khartoum, Nyala, Atbara, Wad Madani and Elobied regions were collected and tested for flavivirus-antibodies by blocking ELISA and confirmed the presence of WNV antibodies 64.81% (477/736*100); 102 (94.44%) of 108 sera from naturally exposed horses and in 229(88.75%) of 260 sera from donkeys and 146(39.67%) of 368 sera from chicken. The highest prevalence was recorded in Khartoum locality and the lowest prevalence was demonstrated in Elobied locality. These results provide evidence of the circulation of WNV in horses, donkeys and chickens in different states in Sudan. This paper present seroprevalence investigation of WNV in horses, donkeys and chickens for the first time in Sudan. Further research study should be taken in the Spread, transmission, isolation and control of the disease.

Keywords: West Nile Virus; Serology; Horses; Donkeys; Chickens; ELISA; Sudan

Introduction

West Nile fever (WNV) is a mosquito-borne viral disease that can affect birds, humans and horses causing inapparent infection, mild febrile illness, meningitis, encephalitis, or death (OIE 2018). WNV is a member of the Flaviviridae family in the genus *Flavivirus*.

It was first isolated from the blood of a woman with a febrile disease in the West Nile district of Uganda in 1937 (Smithburn et.al.1940) [1]. Since then, the virus was circulated in several East African and Asian countries, Kenya, South Africa, Madagascar, Thailand, India (Hayes et.al. 2005) [2].

Therefore, laboratory based diagnostic techniques such as detection of the viral genome by real-time RT-PCR (Reverse-Transcriptase- Polymerase Chain Reaction) or detection of WNV-specific IgM or IgG antibodies by ELISA are useful for detecting any acute WN infections in equids (Castillo-Olivares & Wood 2004) [3]. Infected birds with viremia transmitted the virus to mosquito. 300 bird species may act as vertebrate hosts for WNV (Greene & Reid 2013) [4] and infected migratory birds are thought to spread the viruses to wild birds living in disease free areas (Greene & Reid 2013) (koomar et .al 2003) [4,5]. Infected birds develop a high-titer viremia that allows transmission to feeding mosquitoes, particularly those belonging to the genus *Culex*, which are considered the principal vectors of WNV. Most infected birds usually survive WNV infection, but certain species have been shown to develop fatal disease (Venter. et.al 2010) (Campbell et.al 2002) [6,7]. Among clinically diagnosed humans and horses, mortality rates can be up to 10% and 25–45%, respectively (Greene & Reid 2013) (Epp et.al. 2007) [4,8].

In Sudan the first outbreak of WNV in human reported in children aged up to 14 years from the Nuba Mountains in 1956 (Taylor et.al. 1956) [9]. (Evelyn Depoortere et, al) [10] confirmed outbreak of West Nile virus in children from Nuba Mountains in 2002, Yousof et.al 2018 [11] reported the present of anti-WNV IgG antibody 40 (44.4 %) in blood human sera, and 2 (2.2%) were reactive for anti-WNV IgM antibody. West Nile Virus detected, inside wild mosquitoes in Khartoum capital of Sudan using PCR, (Ali .et, al 2020) [12]. No data was found about the disease situation in animal in the Sudan.

In the present study we attempted to investigate the natural circulation of WNV by serological tests in equines and chickens in five Regions in Sudan, more studies in the disease such

as monitoring of geographic spread and dynamics of WNV transmission in both primary and accidental hosts, isolation and control will be recommended.

Materials and Methods

Blood samples (736) were collected from the jugular vein of healthy horses, donkeys and chickens unvaccinated against WNV, (108) horses, (260) donkeys and (368) chickens, centrifuged and stored at -20°C until being processed. Samples from horses and donkeys were obtained between October, 2015, and November, 2016 and samples from chickens were collected during 2018–2021. All samples from different localities in five States, Atbra (River Nile State) Khartoum (Khartoum State) WadMadani (Algaeera Stat) Elobeid (North Kurdofan State) Nyala (South Darfour State) (Figure 1).

The collected sera from horses and donkeys were tested using the ID Screen West Nile Competition Enzyme-Linked Immunosorbent Assay (cELISA); IDVET, France which detect antibody directed against (pr-E) envelope WNV protein. Chickens sera were performed by INGEZIM WestNile Compac, Spain, it is blocking immunoenzymatic assay for the specific detection of antibodies to west Nile virus in birds and equine serum samples. The test was performed in accordance to the manufacturer's instruction. Results obtained were statistically analyzed using Statistical Packages for Social Science (SPSS) version 16, Software. The statistical significance between infection and region was determined using frequency and the chi-square analysis.

Result and Discussion

The prevalence of (WNV) antibodies in the total sera samples examined (736) using Competition Enzyme-Linked Immunosorbent Assay (cELISA) was 64.81% ($477/736*100$); regarding the species: Antibodies against (WNV) were detected 94.44($102/108*100$) in horse sera, 88.07% ($229/260*100$) donkey and 39.67($146/368*100$) chicken samples (Tables 1 and 2). The highest prevalence (100%) was detected in horses sampled from Nyala and the lowest prevalence (07.25) was recorded in chickens sampled from Elobied. The prevalence of the WNV antibodies in donkeys sampled is between the value of 97.87 ($46/47*100$) in Khartoum and 80.00($44/55*100$) in WadMadani (Table 1) while that the highest prevalence of the disease in Chicken sera was reported in WadMadani 55.56 ($35/63*100$), and the lowest 07.25 ($05/69*100$) in Elobied locality (Table 2).

Across the regions; Khartoum locality showed highest prevalence 74.85 (125/167*100) followed by Nyala 69.19.85(128/185*100), WadMadani, 68.03 (83/122*100), Atbra 66.90(95/142*100) and the lowest prevalence was reported in Elobied locality 38.33 (46/120*100) (Table 3). Statistically, the chi-square analysis showed that; there is an association between infection and location (P = 0.001).

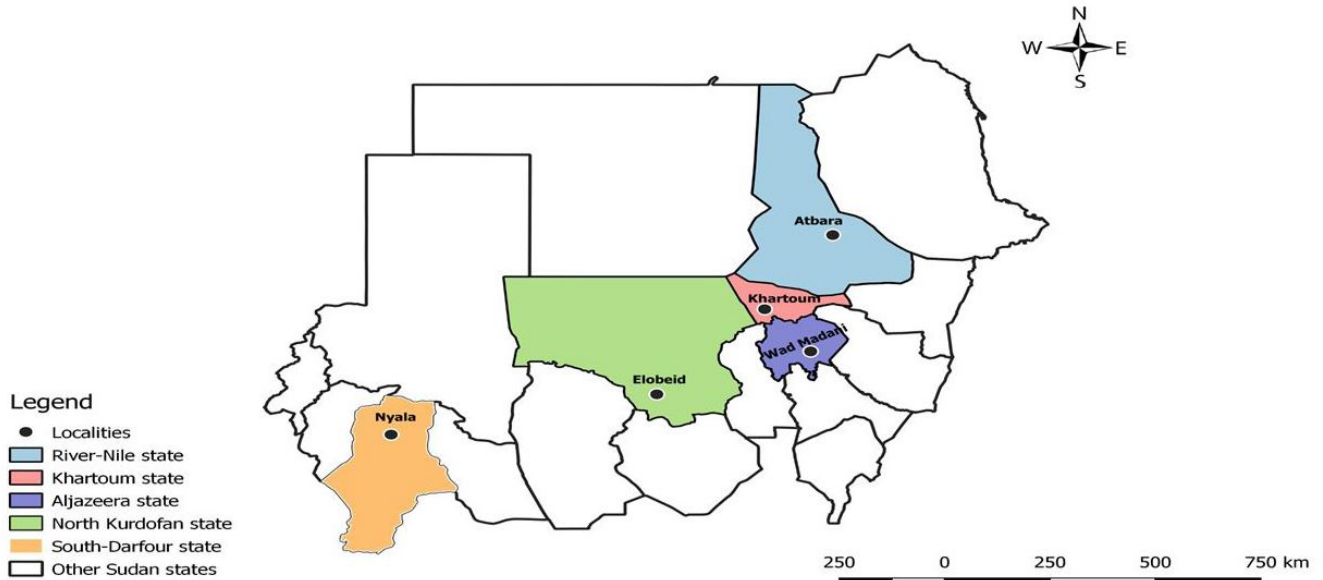


Figure 1: Localities of serological investigation of (WNV) antibodies in Equids and Chickens sera in localities at five States of the Sudan

Localities	Date of samples collection	Horses' Samples Examined	NO. of positive samples	NO. of negative samples	Prevalence %	Donkeys' samples examined	NO. of positive samples	NO. of negative samples	Prevalence %
Khartoum	2015-2016	45	39	6	86.66	47	46	01	97.87
Nyala	2015-2016	53	53	0	100	40	33	07	82.5
Atbara	2015-2016	06	06	0	100	67	65	02	97.01
Wad-Madni	2015-2016	04	04	0	100	55	44	09	80.0
Elobied	2015-2016	00	00	0	0	51	41	10	80.39
Total	0	108	102	6	94.44	260	229	29	88.07

Table 1: Prevalence of West Nile virus in Horses and Donkeys

Localities	Date of samples collection	Total samples examined	NO. of positive samples	NO. of negative samples	Prevalence %
Khartoum	2018-2020	75	40	35	53.33
Nyala	2018-2020	92	42	50	45.65
Atbara	2019-2021	69	24	45	34.78
Wad-Madni	2019-2021	63	35	28	55.56
Elobied	2019-2021	69	05	64	07.25
Total		368	146	222	39.67

Table 2: Prevalence of West Nile virus in Chickens

Localities	Horses examined	Donkeys examined	Chickens examined	Total samples examined	No. positive	Prevalence%
Khartoum	45	47	75	167	125	74.85
Nyala	53	40	92	185	128	69.19
Atbara	06	67	69	142	95	66.90
Wad-Madani	04	55	63	122	83	68.03
Elobied	00	51	69	120	46	38.33

Table 3: Prevalence of WNV in five localities in the Sudan

This investigation reports the first detection of WNV circulation in equids and chickens populations 64.81% ($477/736 \times 100$); Seropositive were detected in the localities of five States in Sudan. The seroprevalence rate in equids samples reported in this study 89.95 ($331/368 \times 100$) is higher than the rate of chicken samples 39.67 ($146/368 \times 100$) our result is similar to previous studies conducted in Poland (Bazanów et.al 2018) [13] who showed that serum neutralizing antibodies to WNV is 5 (35.7%) of 14 birds and 62 (15.08%) of 411 horses, respectively, our result also agree with (Eyboosh. et.al 2019) [14] who mentioned that The highest seropositivity rate was observed among equids 100% in Morocco and highest seroprevalence among birds was seen in Tunisia 23% and this results was explained by (Komar. 2001) [15] (Komar et.al 2003) [5] who explored that domestic birds like chickens do not develop sufficient viremia to permit a transmission cycle and so are considered as dead end hosts.

Despite of limited number of horses samples in Atbara and WadMadani localities and no samples from Elobied locality, due to low populations of horse in country comparing with the populations of donkeys, horses samples presented a highest seroprevalence rate detected 94.44 ($102/108 \times 100$) and this is agree with work study conducted in Senegal Valley confirmed high prevalence rates (85%, $n = 367$). (Chevalier et al., 2010) [16]. Similar findings were also reported earlier in South Africa (75% in mares, $n = 243$) (Guthrie et al., 2003) [17]. From current study we observed lower seropositive in Donkeys samples 88.07% ($229/260 \times 100$) comparing with horse samples 94.44 ($102/108 \times 100$) Similar findings demonstrated by (Azmi et al. 2017) [18] Seroprevalence in horses (82.6%) was significantly higher than in donkeys and mules (39. 3%) and (Lafri et al., 2017) [19] who mentioned that about 26% of the tested horses and 14% of the tested donkeys had antibody to WNV (Lafri et al., 2017) [19].

On the other hand, Khartoum locality reported high rate 74.85 ($125/167 \times 100$) comparing with other localities and this may be due to expose horse to the virus through mosquito bites, (Ali et.al 2020) [12] recorded West Nile Virus inside wild mosquitoes in Khartoum capital of Sudan using PCR [20].

In our study, we concluded that high rate of sero prevalence of West Nile virus among equids and chickens in the Sudan.

Declarations

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Author Contributions

Wegdan H Ali designed the study and wrote the paper; Wegdan,H.A, Rayan,M.A, Muzdalifa.A.H. , Sana,I. Mohamed analyzed the data.,Rayan, M.A. ¹, Shaza, M.M. ¹, Salma,O.A, Muzdalifa.A.H. ,Sana, I. Mohamed performed the laboratory work.,Wegdan, H. Ali, Rayan, M.A. , Sana, I. Mohamed. and Saafass, M.A. Alsarraj² contributed reagents/materials /Samples.

Statement of Animal Rights

Our study did not include laboratory experiment in animal

Conflict of Interest Statement

The authors declare that we have no conflict of interest.

Data availability statement

The datasets generated during the current study are available from the Journal and the corresponding author on reasonable request.

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