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Sero Prevalence of Equine Infectious Anemia (EIA) Virus in Selected Regions in Sudan

H. A. Wegdan[1*], M. E. Sahar[1], A. Ballal[2], K. S. Intisar[3], M. M. Shaza[1], O. A. Algezoli[2], H. A. Ihsan[2], A. M. Baraa[1], K. M. Taha[4], E. M. Nada[5], A. A. Manan[2], Y. H. Ali[3] and Y. M. Nouri[6]

[1]Department of Virology, Veterinary Research Institute, Animal Resources Research Corporation, P.O.Box 8067, ALAmarat, Khartoum, Sudan.
[2]Department of Viral Vaccines, Veterinary Research Institute, Animal Resources Research Corporation, P.O.Box 8067, ALAmarat, Khartoum, Sudan.
[3]Department of Biology, Faculty of Science and Arts, Northern Border University, Rafha, Kingdom of Saudi Arabia.
[4]Atbara Veterinary Research Laboratories, P.O.Box 121 Atbara, River Nile State, Sudan.
[5]Wad Medani Veterinary Research Laboratories, P.O. Box 555, Gezira State, Sudan.
[6]ElObied Veterinary Research Laboratories, P.O.Box 373, North Kordofan State, Sudan.

Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

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ABSTRACT

Equine infectious anaemia (EIA) is an acute to chronic lentivirus disease affecting members of equidae. In this study, the prevalence of EIA virus antibodies was investigated in 358 sera samples collected randomly from apparently healthy horses and donkeys during 2008-2013. Seven regions in Sudan were investigated, including Khartoum, Nyala, Atbra, Elfashir, Halfa, Madani and Kurdofan. Results revealed that, the prevalence of EIA virus antibodies in the total samples examined using

**Corresponding author: E-mail: wegdanhassan@hotmail.com;*

Indirect ELISA, was 5.58% (20/358*100); of which 3.17% (6/189*100) horse samples and 8.28% (14/169*100) donkey samples. Across the regions sampled; the highest prevalence (11.1%) was detected in donkeys from Halfa. However, all of the sampled donkeys and horses from Atbara were sero-negative to EIA virus.

Conclusion: Equine Infectious Anemia Virus was detected in horses and donkeys in selected regions in Sudan.

High sero-prevalence of the disease in horse from Elfashir and donkeys from Nyala was recorded. The highest prevalence in regions examined was detected in Halfa. Statistically, the chi-square analysis showed that; there is an association between infection and location (P = 0.001).

Further studies for virus and antibody detection such as PCR and ELISA should be conducted to enables rapid identification of Equine Infectious Anemia Virus and to monitor the prevalence of the disease in other parts of the country.

Keywords: Equine; infectious anaemia; sero-prevalence; horse; donkeys; ELISA; Sudan.

1. INTRODUCTION

The causative agent of equine infectious anemia (EIA) virus is a*lentivirus* in the family *Retroviridae* of subfamily *Orthoretrovirinae* [1]. The disease is prevalent in all parts of the world and it is of importance for the equine industry [2]. Infection with the virus is characterized by recurrent febrile episodes with high virus load and anemia. Recovered animals may suffer from recurring clinical (chronic) disease caused by new mutant EIA virus strains, which is characterized by irregular and less frequent viremia, associated with weight loss, dependent edema, progressive non regenerative anemia [3,4,5] and are often termed as a “swamper” [6]. The disease then, progresses to an asymptomatic, but, still infectious stage which remains for the rest of the animal's life [7], and antibody-positive animals older than 6–8 months are identified as virus carriers [8].

The disease could be transmitted by transferring of virus-infective blood by blood-feeding insects between horses in close proximity. The virus is found free in the plasma or cell associated, principally in monocytes and macrophages of infected animals [9]. Transmission of EIA by biting flies is purely mechanical, additionally; EIA virus could be readily transmitted iatrogenically through the use of blood-contaminated syringes, needles, or surgical equipment, or by transfusion of infective blood or blood products [9].

For diagnosis, agar gel immunodiffusion (AGID) tests [10] and enzyme-linked immunosorbent assays (ELISAs) [11,12] are accurate, reliable tests for the detection of EIA in horses, except for animals in the early stages of infection and foals of infected dams [13]. AGID test [14] and ELISA which uses purified viral particles as antigen [15] [16] or recombinant EIA virus proteins [17,18,19], are used as screening test and in surveillance studies for EIA virus in recent years [20,21] .

Although EIA virus has a worldwide distribution [8,22]; however, there is no enough information about the disease situation in Sudan. The only study conducted in Khartoum state of Sudan [23] cannot be considered as a true reflection of the disease in Sudan. It is important to know the situation in other regions of the country; therefore, the aim of this study is to investigate the presence of antibodies against EIA virus in horses and donkeys in selected regions in Sudan.

2. MATERIALS AND METHODS

2.1 Samples

A total of 358 blood samples were randomly collected (apparently healthy or diseased animals) from horses and donkeys from seven regions in Sudan during 2008-2013; including, Khartoum, Nyala, Atbra, Elfashir, Halfa, Madani and Kurdufan (Table 1). Following collection and blood clot, Sera were separated, clarified by centrifugation and stored at -20℃ until they were tested for anti- EIA virus antibodies using ELISA.

2.2 ELISA Test

EIA virus ID Screen kit (ID Vet Company, France), an indirect ELISA for the detection of p26 (GAG gene) antibodies directed against the EIA virus in horse serum was used. The test was performed according to the manufacturer’s instruction.

2.3 Statistical Analysis

Data was inserted into Microsoft Office Excel (2010) spread sheet program (Microsoft

Corporation) to create a database and transferred to Statistical Packages for Social Science (SPSS) version 16, Software. The statistical significance between infection and locality was determined using frequency, mean, cross tabulation and the chi-square analysis.

3. RESULTS

The prevalence of EIA virus antibodies in the total sera samples examined using an indirect ELISA was 5.58% (20/358*100); of which 3.17(6/189*100) horse samples and 8.28% (14/169*100) donkey samples (Tables 2, 3 & 4). Across the regions sampled; the highest prevalence (11.1%) was detected in donkeys sampled from Halfa. However, all of the sampled donkeys and horses from Atbara were sero-negative to EIA virus.

Statistically, the chi-square analysis showed that; there is an association between infection and location (P = 0.001).

4. DISCUSSION

In the current study, the prevalence of antibodies against EIA virus was studied in horses and donkeys from different regions in Sudan; including, Khartoum, Nyala, Atbra, Elfashir, Halfa,Madani and Kurdufan.

Using an indirect ELISA, in total of 358 serum samples examined, the prevalence of EIA virus antibodies was 5.58% (20/358*100) of which 3.17% (6/189*100) horse samples and 8.28% (14/169*100) donkeys sample. The highest prevalence of the disease in different regions was detected in Halfa. When comparing species, in different regions examined, horses from Elfasher and donkeys from Nyala showed the highest prevalence 5% & 15.3% respectively. This result is higher than that obtained by Ibrahim [23] in donkeys from Khartoum state and that of Getacheus et al. [22] who found 0.2% prevalence out 1002 sera sample tested in Central Ethiopia. However, similar studies conducted in UAE [24], Bulgaria [25] showed absence of the disease in donkey populations; and in donkey and horse in Turkey [26].

The variation in the prevalence rate obtained for donkeys and horses in this study is justifiable considering donkeys population in Sudan to be 7506000 [27] which is also 10 times the horses population; their distributed all over the country and their uses in different activities, like transport and farming. Horses on the other hand are restricted with larger numbers in some regions where they are considered economically important based on their uses in racing, and transportation."

In our study, antibodies to EIA virus were not detected in all samples collected from Atbara; although Atbara is not far away from Khartoum where the disease was previously detected with a prevalence of 8.7% [23], which is a bit higher than we found (2.2% in horses and donkeys) in this study. For other regions in Sudan, no previous serological or virological studies were conducted concerning EIA. A wide range of laboratory tests are available that allow more rapid diagnosis of EIA. AGID and ELISA are used by many researchers [28,29] for diagnosis of the disease; but they could not detect the disease in the early stage of infection and in foals of infected dams [30,11]. ELISA test has been used by different laboratory and field studies [31,12], with high sensitivity and specificity.

Serological evidence of EIA virus infection has been reported in many countries around the world, Serbia 0.17% [32], Brazil 0.57 % [33], Italy 17.5 % [34]; but the disease was not reported in African countries till 2009 [35], followed by serological report of the disease in Nigeria in 2014 [36] and other African countries such as; Ethiopia [22] and Central African Republic; Chad, Egypt, Eritrea, Libya and South Sudan [37].

Table 1. Region distribution of sampled horses and donkeys in Sudan between 2008 and 2013

Region	No of horses	No of Donkeys	Total (equidae) samples
Khartoum	89	-	89
Nyala	63	39	102
Atbra	24	30	54
Elfashir	40	-	40
Halfa	-	27	27
Madani	-	18	18
Kurdofan	-	28	28
Total	189	169	368

Table 2. The prevalence of EIA virus in different regions in Sudan

Region	Total no. of samples	Number positive	% prevalence
Khartoum	89	2	2.2%
Nyala	102	9	8.8%
Atbra	54	0	0%
Elfashir	40	2	5%
Halfa	27	3	11.1%
Madani	18	1	5.6%
Kurdofan	28	3	10.7%

Table 3. The prevalence of EIA virus in horses

Region	Horses		
	+ve	-ve	% prevalence
Khartoum	1	61	1.6
Nyala	3	60	4.76
Atbra	0	24	0
Elfashir	2	38	5
Halfa	0	0	0
Madani	0	0	0
Kurdufan	0	0	0
Total	6	183	3.17

Table 4. The prevalence of EIA virus in donkeys

Region	Donkeys		
	+ve	-ve	% prevalence
Khartoum	1	26	3.7
Nyala	6	33	15.3
Atbra	0	30	0
Elfashir	0	0	0
Halfa	3	24	11.1
Madani	1	17	5.5
Kurdufan	3	25	10.7
Total	14	155	8.28
Total of samples (horses & donkeys) =358		+ve=20%	prevalence =5.58

5. CONCLUSIONS

Equine Infectious Anemia Virus was detected in horses and donkeys in Sudan. High sero-prevalence of the disease in horse from Elfashir and donkeys from Nyala was recorded. The highest prevalence in regions examined was detected in Halfa. Statistically, there is an association between infection and location.

Further studies for virus and antibody detection such as PCR and ELISA should be conducted to enables rapid identification of Equine Infectious Anemia Virus and to monitor the prevalence of the disease in other parts of the country.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. OIE, Terrestrial Manual. Equine infectious anaemia. Chapter 2.5.6; 2013.
Available:http://www.oie.int/fileadmin/Home/fr/Healthstandards/tahm/2.05.06_EIA.pdf
2. Motie A. An outbreak of suspected equine infectious anemia in Guyana. British Veterinary Journal. 1986;142:36–40.

3. Sellon DC. Equine infectious anaemia. Vet Clin North Am-EqPrac. 1993;9:321–335.
4. Riegel CM, Stockham SL. Anemia associated with bacterial and viral infections. In: Schalm's Veterinary Hematology, 6th Ed, Douglas JW and KJ Wardrop (eds). Blackwell Publishing Ltd. Ames, Iowa, USA. 2010;213-214.
5. Hammad M Hussain, Ghulam Muhammad, Ijaz Javed, Muhammad Siddique. Sero-survey of equine infectious anemia in five draught equine populated metropolises of Punjab, Pakistan. Pak. Vet. J. 2012;32(2): 247-250.
6. Clabough DL, Gebhard D, Flaherty MT. Immune-mediated thrombocytopenia in horses infected with equine infectious anemia virus. J Virol. 1991;65:6242-6251.
7. Hammond SA, Cook SJ, Lichtenstein DL, Issel CJ, Montelaro RC. Maturation of the cellular and humoral immune responses to persistent infection in horses by equine infectious anemia virus is a complex and lengthy process. J. Virol. 1997;71:3840-3852.
8. Manual of Diagnostic Tests and Vaccines for Terrestrial Animals; 2015. Available:http://www.oie.int/international-standard-setting/terrestrial-manual/access-online
9. Available:http://www.merckvetmanual.com/mvm/index.jsp
10. Coggins L, Norcross NL, Nusbaum SR. Diagnosis of equine infectious anaemia by immunodiffusion test. American Journal of Veterinary Research. 1972;33:11–17.
11. Suzuki T, Ueda S, Samejina T. Enzyme linked immunosorbent assay for diagnosis of equine infectious anaemia. Veterinary Microbiology. 1982;7:307–316.
12. Cullinane A, Quinlivan M, Nelly M, Patterson H, Kenna R, Garvey M, Gildea S, Layons P, Flynn M, Galvin P, Neylon M, Jankowska K. Diagnosis of equine infectious anemia during the 2006 outbreak in Ireland. Vet Rec. 2007;161: 647-652.
13. Toma B. Réponse sérologique négative persistante chez une jument in-fectée. Recueil de Medecine Veterinaire. 1980; 156:55–63.
14. Coggins L, Norcross N. Immunodiffusion reaction in equine infectious anemia. Cornell Vet. 1970;60:330–335.
15. Burki F, Rossmanith W, Rossmanith E. Equinelentivirus: Comparative studies on four serological tests for the diagnosis of equine infectious anaemia. Vet. Microbiol. 1992;33:353–360.
16. Pimenta dos Reis JK, Leite RC. An enzyme linked immunosorbent assay (ELISA) test for the diagnosis of equine infectious anemia in Brazil. Prev. Vet. Med. 1994;20:261–267.
17. Birkett AJ, Ye´lamos B, Rodriguez-Crespo I, Gavilanes F, Peterson DL. Cloning expression, purification, and characterization of the major core protein (p26) from equine infectious anemia virus. Biochim. Biophys. Acta. 1997;1339:62–72.
18. Kong XG, Pang H, Sugiura T, Matsumoto Y, Onodera T, Akashi H. Evaluation of equine infectious anemia virus core proteins produced in a baculovirus expression system an agar gel immunodiffusion test and enzyme-linked immunosorbent assay. J. Vet. Med. Sci. 1998;60:1361–1362.
19. Adriana S Toledo Piza, Alessandra Rael Pereira, Maria Thereza Terreran, Otto Mozzer, Amı´lcar Tanuri, Paulo Eduardo Branda˜o, Leonardo Jose´ Richtzenhain. Serodiagnosis of equine infectious anemia by agar gel immunodiffusion and ELISA using a recombinant p26 viral protein expressed in *Escherichia coli* as antigen. Preventive Veterinary Medicine. 2007;78: 239–245.
20. Gill FR, Hussain MH, Muhammad G, Mansoor MK, Hafeez MA, Bilal MQ, Tahir U. Enzyme linked immunosorbant assay (ELISA) based seronegative findings on the investigations of equine infectious anemia in Faisalabad Metropolis (Pakistan). In: Proceedings of 29th World Veterinary Congress held on July, 27-31 at Vancouver, Canada; 2008.
21. Albayrak H, Ozan E. Serosurveillance of equine infectious anemia in the Ardahan province of Turkey. Trop Anim Health Prod. 2010;42:1593-1595.
22. Getachew M, Alemayehu F, Chala C, Amare B, Kassa D, Burden F, Wernery R. A cross sectional sero-survey of some infectious disease of working equidis in Central Ethiopia. Jornal of Veterinary Medicine and Animal Health. 2014;231-238.
23. Ibrahim, Gehan Ibrahim Aljak. Sero-prevalence and risk factors of equine infectious anemia (EIA) in Khartoum State –Sudan. Thesis; 2014. Available:http://repository.sustech.edu/handle/123456789/10723

24. Turnbull A, Wernery U, Wernery R, Anandh JP, Kinne J. Survey of six infectious diseases of feral donkeys in the United Arab Emirates. Equine Vet. Educ. 2002;14:33-38.
25. Chenchev IV, Rusenova N, Sandev N. Sero-epidemiological studies of donkeys' blood for detection of some virus infections onungulates. Trakia J. Sci. 2011;9:82-86.
26. Ataseven VS, Arslan HH. Equine infectious anemia in mules, donkeys and horses: Epidemiologic studies in the different geographic regions of Turkey. J. Equine. Vet. Sci. 2005;25:439-441.
27. MARF. Ministry of Animal Resources and Fishers; Statistical bulletin for Animal Resources. No 19- Information Centre; 2010.
28. Ball JM, Henry NL, Montelaro RC, Newman MJ. A versatile synthetic peptide based ELISA for identifying antibody epitopes. Journal of Immunological Methods. 1994;171:37–44.
29. Grund CH, Lechman ER, Pezzuolo NA, Issel CJ, Montelaro RC. Fine specificity of equine infectious anaemia virus gp90-specific antibodies associated with protective and enhancing immune responses in experimentally infected and immunized ponies. Journal of General Virology. 1996;77:435–442.
30. Pearson JE, Knowles RC. Standardization of the equine infectious anemia immunodiffusion test and its application to the control of the disease in the United States. Journal of the American Veterinary Medical Association. 1984;184:298–301.
31. Susan D, Dallas G, Mills H, Burr P. Assessment of 2 commercial ELISAs for EIA and EVA serology. Biobest, UK; 2008.
Available:http://www.biobest.co.uk/assets/files/diagnostics/serologicaltesting.pdf
(Accessed on July 28, 2011)
32. Branka Vidić , Sara Savić, Živoslav Grgić, Dejan Bugarski, Diana Lupulović, Nadežda Prica, Doroteja Marčić. Serosurveillance of equine infectious anaemia in a region of Vojvodina. Arhiv Eveterinarske Medicine. 2014;7(2):3-12.
33. Marlon Cezar Rebelatto, Clóvis de Oliveira, Rudi Weiblen, Saul Fontoura da Silva, Luiz SérgioSegala de Oliveira. Serological Diagnosis of equine infectious anemia vírus infection in the central region of the riogrande do sul state. Ciência Rural. 1992:22(2):191-196.
Available:https://dx.doi.org/10.1590/S0103-84781992000200012
34. Katia Cappelli, Stefano Capomaccio, Frank R. Cook, MichelaFelicetti, Maria Luisa Marenzoni, Giacomo Coppola, Andrea Verini-Supplizi, Mauro Coletti, Fabrizio Passamonti J. Clin. Microbiol. 2011;49(1):27-33.
35. Available:http://www.oie.int/Wahidinterface
36. Mayaki AM, Adeyefa CAO, Aiki-Raji CO. Detection of IgG and/or IgM antibodies against equine infectious anaemia virus (EIAV) in Nigerian race and polo horses. Sokoto Journal of Veterinary Sciences. 2014;12(3):32-36.
37. Available:http://www.oie.int/wahis_2/public/wahid.php/Diseaseinformation/statusdetail

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