Serologic Evaluation of the Equine Infectious Anemia in Kars and Ardahan - Turkey

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Summary

The material consisted of 476 equids including 400 horses and 76 donkeys from Kars and Ardahan provinces, North-Eastern of Turkey. Blood was collected from all horses and donkeys and the sera were analyzed for the presence of antibodies to equine infectious anemia virus (EIAV) using an agar gel immunodiffusion (AGID) test. The results revealed that none of the horses and donkeys had antibodies for EIAV.

Keywords: Equine infectious anemia, Donkey, Horse

Kars ve Ardahan'da Equin Enfeksiyöz Aneminin Serolojik Araştırılması

Özet

Çalışmanın materyalini Kars ve Ardahan illerinden sağlanan 400 at ve 76 eşek olmak üzere toplam 476 tek tırnaklı oluşturdu. Tüm at ve eşeklerden alınan serum örneklerinde equine infectious anemia virus (EIAV) enfeksiyonunun varlığını belirlemek için agar gel immunodiffusion (AGID) testi kullanıldı. Çalışma sonucunda at ve eşekler EIAV yönünden negatif bulundu.

Anahtar sözcükler: Equin enfeksiyöz anemi, Eşek, At

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INTRODUCTION

Equine infectious anemia (EIA) is caused by the equine infectious anemia virus (EIAV), classified in the subfamily Lentivirinae of the family Retroviridae ^{1,2}. Equine infectious anemia (EIA) is a disease that affects equidae (such as horses, mules, donkeys) only ²⁻⁴.

The virus can be transmitted through secretions and excretions of acutely or chronically infected animals, blood transfusions, blood–contaminated materials and blood sucking insects such as horseflies, deer flies and Stomoxys calcitrans^{2,5-7}. The highest number of cases occurs in warm climates because of the prevalence of blood sucking insects that are the primary transmitters of this disease ^{5,8}.

There are acute, chronic, and inapparent forms of the disease ⁹. Classic signs of acute EIA include fever, anemia, icterus, ventral edema, and weight loss. Severe clinically signs of EIA were frequently observed in horses, but not in donkeys or mules ³. Despite the lack of clinical signs, some horses represent a source of infection to other non-infected horses ¹⁰. Animals generally recover from either the acute or chronic form of the clinical disease, but will remain lifelong carriers of the virus. The persistent carriers have played the major epidemiological role in spreading of EIA infection ^{2,4,7,8,11}. The EIA virus specific antibodies in donkeys and horses were identical but horse-adapted strains of EIAV were unable to produce clinical disease in the donkeys tested. Therefore, clinical manifestations of EIA were absent in donkeys because the concentration of virus remained below this critical plasma level than in horses ³.

The agar-gel immunodiffusion (AGID) test, formerly named the Coggins test is commonly used for the diagnosis of EIA ¹². The test is a reference test and is used in many countries ^{8,13,14} during importation and exportation since EIA is on List-B as indicated by the Office International des Epizooties (OIE). Other tests such as enzyme linked immunosorbent assay (ELISA) and competitive ELISA (C-ELISA) have also been introduced for the diagnosis of EIA ^{8,13,15-17}.

Recently, molecular biological techniques have been applied to detect EIAV in tissues and blood 4,11,18.

Studies concerning EIA have been carried out in different parts of Turkey ^{14,19,20} but no report exists in North-Eastern Anatolia.

MATERIAL and METHODS

Blood samples were collected randomly from 400 horses (225 male and 175 females) and 76 donkeys (54 male and 22 female) older than 1 year without clinical signs of the disease from Kars and Ardahan provinces in the North-Eastern part of Turkey between February and June 2007. The blood samples were taken into clot activator vacuum tubes and centrifuged at 3000 rpm for 10 min. The serum samples were inactivated at 56°C for 30 min and were stored at –20°C until use. Horses and donkeys were housed between October and April. Animals were fed hay, barley and straw during indoor period and grazed during outdoor period.

The commercial AGID kits were obtained from VMRD Inc., USA and the test was carried out as described by the manufacturer. Suspected samples were retested by AGID.

RESULTS

In this study, a total of 476 equines (400 horses and 76 donkeys) were tested by AGID. All animals were negative for antibodies against to EIAV.

DISCUSSION

The international trading of equines and semen are restricted due to the contagious diseases. In most of these countries, the EIA infection is a noticeable disease, and the control measures include official screening or monitoring, precautions at borders, control of movement inside the country ^{21,22}.

In present study, all animals were negative for antibodies against to EIAV. Due to the heavy winter conditions and a wide range of uneven lands in Kars and Ardahan district, horses and donkeys still serve an important role in human life participating in transport and agriculture. Hence, it is crucial to minimize the potential risk great economical losses in resulting from infectious diseases occurring in animals. EIA infection is widely reported from the several countries ^{8,11,23}. Kars and Ardahan provinces constitute a great risk for the occurrence of EIA in horses and donkeys because of high population of hematophagous vectors ²⁴ and as the provinces placed on border of Armenia and Georgia. Moreover, high number of uncontrolled animal movements pose another significant threat factor for the spreading the infection to the other parts of Turkey. The North-Eastern Anatolian region of Turkey has border with Armenia, Georgia and control of this border is very difficult given the geographical situation. Movement at the border creates important health risks for the equidae population of North-Eastern Anatolia in Turkey, because we do not know the situation of EIA infection in those countries ^{14,17,19,20,23,25}. A control program in this region based on serological testing would promote the prevention of the disease. The preventive policy would also require a well-coordinated effort by the government and horse owners.

There are three reports on the frequency of EIAV using AGID and ELISA in Turkey at present ^{14,19,20}. Burgu et al.²⁰ studied 294 horses, Ataseven and Aslan ¹⁹ studied 69 mules, 154 donkeys and 408 horses and Turan et al.¹⁴ studied the prevalence of EIA in 404 horses from different regions in Turkey. They found that animals were not seropositive for EIA. However the seropositivity for EIAV was reported in other several countries ^{17,9,18,25-27}.

In this study, horses and donkeys were negative for antibodies to EIAV. The results of this study are similar to those previous studies indicating the absence of the disease in a large number of horses ^{14,20}, and donkeys ¹⁹. This may suggest that the disease is not present in the sampling district or it might be present but couldn't mount on immune response yet. In future, well-designed epidemiological studies based on molecular techniques are needed to definitively determine animals in the incubation period.

This is the first study on the presence of EIA in Kars and Ardahan provinces in the North-Eastern part of Turkey in where the horse and donkey population is high. The current study indicated that EIA might not become a risk potential for working, racing and breeding of equidae in this geographic district of Turkey.

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REFERENCES

- 1. Sellon DC: Equine infectious anemia. Vet Clin North Am: Equine Pract, 9, 321-335, 1993.
- 2. Cook RF, Montelaro RC, Issel CJ: Equine infectious anemia. In, Studdert MJ (Ed): Virus Infections of Equines. Amsterdam, Elsevier, pp. 297-323, 1996.
- 3. Cook SJ, Cook RF, Montelaro RC, Issel CJ: Differential responses of *Equine caballus* and *Equine asinus* to infection with two pathogenic strains of equine infectious anemia virus. *Vet Microbiol*, 79, 93-109, 2001.
- Spyrou V, Papanastassopoulou M, Psychas V, Billinis Ch, Koumbati M, Vlemmas J, Koptopoulos G: Equine infectious anemia in mules: Virus isolation and pathogenicity studies. *Vet Microbiol*, 95, 49-59, 2003.
- 5. MacAllister C, Floyd C: Equine Infectious Anemia. Division of Agricultural Sciences and Natural Resources, Oklahoma State University, Oklahoma Extension Service, F-9124-2. Available on website; http://www.osuextra.com. *Accessed:* 10.08.2008.
- 6. Foil LD, Issel CJ: Transmission of retroviruses by arthropods. *Annu Rev Entomol*, 36, 355-381, 1991.
- 7. Motie A: An outbreak of suspected equine infectious anemia in Guyana. *Br Vet J*, 142, 36-40, 1986.
- 8. Lew AM, Thomas ML, Huntington PJ: A comparison of ELISA, Fast-ELISA, and gel diffusion tests for detecting antibody to equine infectious anemia virus. *Vet Microbiol*, 34, 1-5, 1993.
- 9. Campbell CT, Nusbaum SN: Epidemiological importance of interstate transport of equines infected with equine infectious anemia virus. *J Am Vet Med Assoc*, 198, 1332-1333, 1991.
- 10. Issel CJ, Adams WV, Meek L, Ochoa R: Transmission of equine infectious anemia virus from horses without clinical signs of disease. J Am Vet Med Assoc, 180, 272–275, 1982.
- 11. Nagarajan MM, Simard C: Detection of horses infected naturally with equine infectious anemia virus by nested polymerase chain reaction. *J Virol Methods*, 94, 97-109, 2001.
- 12. Coggins L, Norcross NL, Nusbaum SR: Diagnosis of equine infectious anemia by immunodiffusion test. *Am J Vet Res*, 33, 11-17, 1972.
- 13. **Pearson JE, Gibson CA:** Standardization of equine infectious anemia immunodiffusion and C-ELISA tests and their application to control of the disease in the United States. *Equine Vet Sci*, 8, 46-49, 1988.
- 14. **Turan N, Yılmaz H, Uysal A, Arslan M:** Seronegative findings on the investigations of equine infectious anemia in the Marmara region of Turkey. *Turk J Vet Anim Sci*, 26, 299-302, 2002.
- 15. **Perryman LE, OÕRourke KL, Mason PH, McGuire TC:** Equine monoclonal antibodies recognize common epitopes on variants of equine infectious anemia virus. *Immunology*, 71, 592-594, 1990.
- 16. **Sugiura T, Matsumura T, Fukunaga Y:** Diagnosis of equine infectious anemia by enzyme-linked immunosorbent assay with viral antigen purified by affinity chromotography. *Bull Equine Res Inst*, 23, 42-48, 1986.
- 17. Sugiura T, Kondo T, Matsumura T, Imagawa H, Kamada M, Ihara T: Field application of enzyme-linked immunosorbant assay for screening of equine infectious anemia. *J Equine Sci*, 6, 15-20, 1995.

- Langemeier JL, Cook RF, Cook SJ, Rushlow KE, Montelaro RC, Issel CJ: The application of PCR to the diagnosis and control of equine infectious anemia. Equine Infectious Diseases VII. Proceedings of the Seventh International conference Tokyo, 8th-11th June, p, 299, 1994.
- 19. 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*, 25, 439-441, 2005.
- 20. Burgu İ, Akça Y, Toker A, Alkan F: Atlarda enfeksiyöz anemi'nin serolojik olarak araştırılması. *Ankara Univ Vet Fak Derg*, 36, 123-128, 1989.
- 21. Office International des Epizooties. Animal health policies, evaluation of veterinary services and role of livestock breeders in the surveillance of animal diseases. Available on website; www.oie.int/downld/NDJAMENA.pdf. *Accessed:* 15.08.2008.
- 22. Office International des Epizooties. Listed diseases.

Available on website; www.oie.int/eng/maladies/ en_classification2008.htm?e1d7. *Accessed*: 15.08.2008.

- 23. **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. *J Am Vet Med Assoc*, 184, 298-301, 1984.
- 24. Erdem F: The population dynamic of mosquito larvae/ pupae (Diptera: Culicidae) in Kars Plateau. Master thesis. University of Kafkas, Institute of Natural Sciences, 2007, Kars.
- 25. Issel CJ, Adams WV: Serologic survey for equine infectious anemia virus in Louisiana. J Am Vet Med Assoc, 174, 286-288, 1979.
- Jazbec I: Prevalence and eradication of equine infectious anemia (EIA) in Slovenia. *Pferdeheilkunde*, 12, 565-568, 1996.
- 27. Thomas RJ: Investigation of equine infectious anemia in Queensland using gel diffusion. *Aust Vet J*, 51, 440-442, 1975.