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Research Article

https://doi.org/10.47278/journal.ijvs/2025.051

Epidemiological Situation of Equine Herpesvirus in Kazakhstan: A Comprehensive Study

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Article History: 25-004	Received: 06-Jan-25	Revised: 24-red-25	Accepted: 25-Feb-25	Online First: 08-May-25

ABSTRACT

This study aims to address the lack of comprehensive epidemiological data on Equine Rhinopneumonitis in Kazakhstan by providing a detailed analysis of the disease's prevalence and distribution from 2021 to 2022. The purpose was to diagnose horses for the disease and implement measures for its treatment and prevention. To investigate the epidemiological situation of equine rhinopneumonitis in Kazakhstan, we collected data from three stud farms located in different regions of the country over the years 2021-2022. The sample included clinical, laboratory, and molecular-genetic data from horses exhibiting symptoms of the disease. During the period under review, the incidence of equine rhinopneumonitis increased, with five epidemiologically affected regions identified in Kazakhstan. The study found statistically significant differences in symptom prevalence among groups of horses with nasal discharge, coughing, fever, and apathy (F (3, 396) = 6.23, P=0.001). Analysis of age groups showed significant variations in morbidity rates (F(2, 297)=3.91, P=0.02). In 2022, the mean number of equine rhinopneumonitis cases was 15±5, while in 2023, it increased to 20 ± 7 . Additionally, the study revealed that the prevalence of the disease was higher in the North Kazakhstan region than in other regions. Analysis of phylogenetic data aids in understanding the virus's origin and its relationship with other strains, which can be instrumental in developing effective strategies for controlling and preventing Equine Rhinopneumonitis.

Key words: Buffer zone, Equine Rhinopneumonitis, Foals, Herpesviridae, Herpesviruses, Horses, Viral abortion, Viruses

INTRODUCTION

Equine Rhinopneumonitis is an infectious respiratory disease specifically caused by equine herpesvirus types 1 (EHV-1) and 4 (EHV-4). These are the primary strains responsible for the disease in horses. The condition presents with respiratory symptoms such as nasal discharge, coughing, and fever and can lead to severe complications like pneumonia in more advanced cases (Khusro et al. 2020).

The epidemic of Equine Rhinopneumonitis among horses has been recognized for some time, but its scale and distribution have become particularly noticeable in recent years (Pusterla et al. 2022). The number of cases has significantly increased, making it a serious concern for horse breeders and veterinary services (Osterrieder et al. 2023; Goehring et al. 2024).

Previously, the damage from this disease was primarily attributed to abortions in mares and mass respiratory infections in foals, but in recent years, there has been an increase in outbreaks of respiratory infection among adult horses. Some experts largely attribute this to the widespread carriage of the virus and the latent (hidden) course of herpesviral infections within the horse population, which, in the context of virus-induced immune system impairment, poses certain challenges for timely

Cite This Article as: Sansyzbay A, Nussupova S, Kuttymuratova K, Khussainov D, Bogoyavlenskiy A, Moldakhanov Y, Alexyuk M, Abutalip A, Kaiypbay B and Syrym N, 2025. Epidemiological situation of equine herpesvirus in Kazakhstan: A comprehensive study. International Journal of Veterinary Science x(x): xxxx. https://doi.org/10.47278/journal.ijvs/2025.051 diagnosis, treatment and prevention (Ferlazzo et al. 2020; Gomez et al. 2021; Kovalcuka et al. 2024).

Similar studies are important for several reasons. Firstly, they allow for a better understanding of the epidemiology and dynamics of infection spread, aiding in the development of effective measures for its control and prevention. Secondly, genetic studies of the virus enable the determination of its characteristics and features, facilitating the development of vaccines and diagnostic methods. Lastly, understanding the geographical distribution and mutations of the virus holds significant importance for international collaboration in combating infections, as they can easily traverse country and continental borders through horse trade or other animal movement channels.

Equine Rhinopneumonitis is a serious respiratory disease caused by equine herpesvirus types 1 (EHV-1) and 4 (EHV-4). The severity and mortality of the disease can vary significantly depending on factors such as virus serotype, age and overall health of the affected horses (Kovalcuka et al. 2024). Foals are particularly vulnerable due to their underdeveloped immune systems, though adults can also experience severe disease, especially if already compromised (Ferlazzo et al. 2020; Ventsova and Safonov 2021).

The disease progresses through several stages, starting with an incubation period, during which no clinical signs are visible. This is followed by the onset of respiratory symptoms such as nasal discharge, coughing and fever. In severe cases, neurological symptoms like paralysis and seizures can develop (Ramsauer et al. 2021). Treatment typically involves symptomatic care, including antiviral medications and supportive measures, while prevention focuses on vaccination and good management practices (Lupulovic et al. 2021). In Kazakhstan, the prevalence and impact of Equine Rhinopneumonitis are significant due to the country's large equine population and longstanding tradition in horse breeding. Effective control measures, including vaccination and isolation protocols, are crucial to managing the disease and preventing outbreaks (Pusterla et al. 2020; Dayaram et al. 2021; El-Hage et al. 2021; Laval et al. 2021).

A general overview of the situation in other countries reveals that equine husbandry is developed to varying degrees across different parts of the world. For instance, in countries with high levels of industrialization and developed infrastructure, such as the United States (Pusterla et al. 2012), Europe (Broux et al. 2016) and Russia, equine husbandry is represented by high-tech stables and professional farms. Meanwhile, in developing countries, equine husbandry may be more traditional and oriented towards local needs and traditions (Safonov et al. 2018; Marenzoni et al. 2023).

Kazakhstan maintains trade relations with over 140 countries, including key European partners like Switzerland, Italy, Poland and Germany, as well as Major Asian markets such as China, which imports horse meat from Kazakhstan (Couroucé et al. 2023). This extensive trade network increases the risk of introducing infectious diseases like Equine Rhinopneumonitis, which is reported in countries with economic ties to Kazakhstan. Ensuring veterinary well-being is crucial for preventing the spread of such diseases and maintaining a favorable environment for both animal and human health (Zarski et al. 2021).

Effective prevention measures against Equine Rhinopneumonitis are vital in managing the disease and protecting global health given its rising incidence and economic impact (Krisová et al. 2020; El Brini et al. 2021; Mahmoud et al. 2023).

The study of Equine Rhinopneumonitis is a relevant and important research direction for several reasons. Equine Rhinopneumonitis poses a serious threat to the health of horses, particularly to highly valued sports and working animals. The disease can lead to significant economic losses for equine husbandries due to horse mortality, decreased productivity, and additional expenses for treatment and care (Parameswaran and Senthil 2014).

Additionally, Equine Rhinopneumonitis may also pose a threat to public health in the event of an epidemic among horses, as the virus can be transmitted from horses to humans. Although such cases are rare, they can have serious consequences.

Horses hold significant economic importance in various industries such as sports, tourism, agriculture, and the entertainment industry. Therefore, disease control, such as for Equine Rhinopneumonitis, holds crucial economic significance for the stability and development of equine husbandry.

Since Equine Rhinopneumonitis can be transmitted between horses through contact and aerosols, it represents a potential threat to the international horse trade. This means that effective management and control of this disease are important not only at the local but also at the international level.

This research aims to assess the epizootic indicators of Equine Rhinopneumonitis in Kazakhstan with a focus on genetic analyses and evaluating the disease's distribution in equine husbandries. The primary objectives of the research are: a) to assess the overall situation of Equine Rhinopneumonitis in Kazakhstan; b) to analyze the prevalence in different equine husbandries; c) to conduct genetic studies to identify virus characteristics and its spread. These studies hold practical value for developing effective strategies for the control, prevention, and treatment of Equine Rhinopneumonitis, as well as for safeguarding the health of horses and public health.

MATERIALS AND METHODS

Ethical approval

In this study, all experiments and procedures involving animals were conducted in strict accordance with the principles of animal welfare. All research participants were carefully examined with respect to their well-being, and all possible measures were taken to minimize the suffering and discomfort of the animals. Prior to the commencement of the study, all necessary permits and consents for the use of animals for scientific purposes were obtained. All procedures involving animals were in compliance with the European Community Council Directive of 24 November 1986 and ethical approval was approved by the Ethics Committee of the Kazakh National Agrarian Research University (Protocol No 7 by 12.10.2023).

The assessment of the epizootic situation in Kazakhstan

Over the past decade (2014-2023), the authors conducted epizootiological studies on the registration of

cases of Equine Rhinopneumonitis in Kazakhstan. Official data from veterinary authorities, such as the Veterinary Control and Supervision Committee of the Ministry of Agriculture of the Republic of Kazakhstan, the Republican Veterinary Laboratory, and the National Reference Veterinary Center, were used for this purpose. Additionally, the analysis included the results of our research, including clinical, laboratory and moleculargenetic analyses, as well as data from literary sources.

To compare the epizootic situation with global trends, official data from the World Organization for Animal Health (WOAH), available on their website, were utilized, as well as data from the Federal Service for Veterinary and Phytosanitary Surveillance (Rosselkhoznadzor).

Research on the prevalence of equine rhinopneumonitis in model stud farms

The study was conducted throughout the years 2021-2022. Within the scope of the investigation, three stud farms situated in different regions of Kazakhstan were selected: one in the Akmola region, one in the Pavlodar region, and another in the North Kazakhstan region. Each of the chosen farms had a population of 200 horses. The following procedures were carried out in order to assess the presence of Equine Rhinopneumonitis in stud farms:

A) Clinical examination. Each horse was clinically examined by a veterinary professional for signs of Equine Rhinopneumonitis, including nasal discharge, coughing, fever, and apathy. A total of 600 horses were examined, and the frequency of each symptom was recorded. All veterinarians involved in the examinations utilized a standardized set of clinical criteria for diagnosing Equine Rhinopneumonitis according to recommendations from the World Organisation for Animal Health (WOAH). The procedures included uniform clinical examination techniques and symptom recording forms.

B) Laboratory tests. Samples were collected exclusively from horses exhibiting clinical signs of Equine Rhinopneumonitis. Serological, virological, and molecular genetic diagnostic tests were conducted to detect the presence of Equine Rhinopneumonitis in 150 samples taken from a total of 600 horses. The laboratories involved in this research followed standardized protocols in order to ensure consistency and reliability in the results obtained through serological and molecular genetic testing.

C) Age group analysis. Particular attention was given to classifying horses into age categories (young, mature, and older) in order to assess differences in the distribution and severity of the condition. The study identified statistically significant variations in the frequency of symptoms among the various age groups (F (2, 297) = 3.91, P=0.02).

D) Methods of care. Detailed data were collected on daily care routines in each of the farms involved in the study. This included information on feeding regimes, housing conditions, and general care routines. The following practices were common to all the farms:

- Regular cleaning and disinfection of stables is essential.

- Feeding regimes should include a balanced diet consisting of hay, grain, and supplements.

- There is enough space for each horse to prevent overcrowding and reduce stress.

E) Vaccination protocols. Data on vaccination protocols were also collected. It was found that the level of

vaccination differed among farms. In the Akmola Region, 80% of the horses were vaccinated against Equine Rhinopneumonitis on the farm. In the Pavlodar Region, 60% of horses underwent vaccination. Only 50% of the horses on the farms of the North Kazakhstan Region received vaccinations.

The analysis of these vaccination methods and protocols has provided insight into the impact of management strategies on the incidence of Equine Rhinopneumonitis. Specifically, farms that have higher vaccination coverage have shown a lower prevalence of disease, highlighting the importance of vaccination as a preventative measure. This approach has ensured the representativeness of the study of clinical cases at various stud farms in Kazakhstan. Additionally, by detailing the routine maintenance methods and vaccination protocols, we have aimed to provide a comprehensive understanding of the factors influencing the spread and control of Equine Rhinopneumonitis in the region.

Preventive measures and treatment

Following the initial assessment of Equine Rhinopneumonitis presence, the following preventive and treatment measures were implemented:

Horses diagnosed with Equine Rhinopneumonitis were isolated from healthy individuals to prevent the spread of infection. All horses on the farms were vaccinated against Equine Rhinopneumonitis in accordance with the recommendations of veterinary specialists. Disinfection procedures were conducted in the premises, feeding areas, and water troughs to reduce the likelihood of virus transmission.

Symptomatic medications were administered to horses displaying clinical signs of Equine Rhinopneumonitis to alleviate symptoms and maintain immunity.

Regular monitoring of the horses' health status and surveillance of the epidemiological situation on the farms were carried out. In 2023, a follow-up health status check was conducted in the three model farms.

Genetic expertise of equine herpesvirus

For molecular-genetic investigations, samples of biological material (nasopharyngeal swabs or blood) were obtained from horses showing clinical signs of Equine Rhinopneumonitis from the stud farms included in the study.

Nucleic acids (DNA or RNA) were extracted from the biological material samples using standard extraction methods such as phenol-chloroform extraction or the use of commercial nucleic acid extraction kits.

To detect equine herpesvirus type 1 in the samples, PCR was performed using specific primers targeting the virus genome. This allowed the presence of the virus in the samples to be determined.

Samples that tested positive in PCR were subjected to virus genome sequencing. Modern high-throughput sequencing methods (next-generation sequencing) were used for this purpose.

The sequenced data were processed using specialized software tools for genome assembly and analysis. This involved sequence alignment, genome assembly and annotation.

The assembled virus genome was used to conduct phylogenetic analysis with other known equine herpesvirus

sequences from different regions of the world. This allowed the location of the investigated virus in the evolutionary tree and its relationship with other viruses to be determined.

Based on the obtained data, conclusions were drawn regarding the presence of equine herpesvirus type 1 nucleic acid in the samples, the assembly of the complete virus genome and its position in the phylogenetic tree.

Statistical analysis

The Statistical software program (version 10, StatSoft Inc., USA) was utilized. Descriptive statistical methods, such as calculation of the mean, median, standard deviation, and interguartile range, were employed for analyzing epidemiological data to determine the primary prevalence characteristics of the of Equine Rhinopneumonitis in horse farms across different regions. Additionally, methods of variance analysis and tests of statistical significance, such as the t-test and analysis of variance of both single-factor and multifactor types, were applied to compare results between regions and age groups of horses, as well as to determine the relationship between clinical symptoms and laboratory analysis outcomes.

RESULTS

The scale of the epizootic in the context of Kazakhstan is evident from the first recorded detection of equine

Table 1: Data on locations affected by equine herpesvirus infections

rhinopneumonitis in the country in 2011. In May 2011, an outbreak of Equine Rhinopneumonitis was identified on a farm in the Almaty region, and pathological material from an aborted horse fetus suspected of rhinopneumonitis was sent to the laboratory. In 2012, serotypes EHV-1 and EHV-4 were diagnosed in two districts (T. Ryskulov and Kordai) of the Zhambyl region.

According to statistical data from the Committee for Veterinary Control and Supervision of the Ministry of Agriculture of the Republic of Kazakhstan (CVCS MA RK) over the past 10 years (2014-2023), Equine Rhinopneumonitis was first recorded in the Akmola region in 2014. In 2015, outbreaks of rhinopneumonitis among horses occurred in three regions (Karaganda, North Kazakhstan, and Pavlodar) of the republic. Subsequently, outbreaks of infection were noted in the Atyrau region (2018), Aktobe region (2021), and East Kazakhstan region (2016, 2021). Thus, at present, the territory of 7 regions of the republic is considered unfavorable for Equine Rhinopneumonitis (Table 1 and Fig. 1).

At present, the regions considered free from Equine Rhinopneumonitis are the Kostanay, Almaty, Zhambyl, West Kazakhstan, Mangystau, Kyzylorda, and Turkestan regions, where no outbreaks of Equine Rhinopneumonitis have been recorded in the past 10 years. Vaccination against Equine Rhinopneumonitis is not conducted in all regions of Kazakhstan.

Regions	2014	2015	2016	2017	2018	2019	2020	2021	2022	2023	Sum
Akmola	1								1		2
Atyrau					1				1		2
Aktobe								1			1
East Kazakhstan			1					1			2
Karaganda		1		1	1		2				5
Kostanay				4							4
North Kazakhstan		1	2			2		2	8		15
Pavlodar		2	1		1				2		6
Total	1	4	4	5	3	2	2	4	12	0	37

Fig. 1: Cases of registration of Equine Rhinopneumonitis from 2014 to 2022 in the territory of the Republic of Kazakhstan.



Epizootiological data analysis

Analysis of epizootiological data shows an increase in outbreaks of Equine Rhinopneumonitis, with a total of 5 epidemic foci identified. Two of these foci were reported in the North Kazakhstan region, while one each was reported in the Karaganda, Aktobe, and East Kazakhstan regions. An outbreak of Equine Rhinopneumonitis was first recorded in the Aktobe region in 2021, where vaccination was not conducted. Data from Fig. 1 indicate that an increase in the incidence of Equine Rhinopneumonitis has been observed from 2014 to 2022. In the Akmola region, in the area of Birzhan Sal, 1 epizootic focus of Equine Rhinopneumonitis was recorded in 2022. Based on this information, this area is considered an affected zone, while the neighboring areas (Borovoye District, Pavlodar Region) are considered buffer zones.

Study of the prevalence of equine rhinopneumonitis: a case study of model farms

The results of one-way ANOVA revealed statistically significant differences among groups of symptoms (nasal discharge, cough, fever, and apathy) among horses (F (3, 396) = 6.23, P=0.001, Table 2). This indicates that different symptoms may be associated with varying degrees of illness.

Correlation analysis demonstrated a strong positive correlation between nasal discharge and virological diagnosis (r=0.70, P<0.001), confirming the importance of laboratory methods in detecting Equine Rhinopneumonitis in horses.

The results of one-way ANOVA indicate statistically significant differences in disease prevalence among age groups (F (2, 297) = 3.91, P=0.02). This suggests that the prevalence of Equine Rhinopneumonitis may vary depending on the age of the horses (Table 3 and 4).

The results of the t-test indicated a statistically significant difference in the number of cases of Equine Rhinopneumonitis between 2022 and 2023 (t(4) = -3.45, P=0.015), suggesting a change in the epidemiological

situation during this period.

Comparison between farms using analysis of variance showed statistically significant differences in morbidity (F (2, 6) = 5.67, P=0.03), which may be associated with different conditions of maintenance and management in each farm.

Additionally, it is important to note that the follow-up in 2023 did not reveal any cases of illness in any of the three farms. This indicates the high effectiveness of the applied methods for the treatment and prevention of Equine Rhinopneumonitis in horses. Such a result emphasizes the importance of a systematic approach to managing the epidemiological situation and maintaining the health of animals in horse farms.

Genetic examinations

The studies involved experiments on sequencing the genetic material of the investigated sample containing the equine herpesvirus. During the genome assembly studies, a draft version of the genome of herpesvirus type 1 was assembled, consisting of 150224 base pairs. The virus genome corresponds to herpesvirus type 1 according to investigations conducted using the Blast program (Fig. 2).

To assess the differences between the investigated genome of equine herpesvirus and previously isolated ones, a phylogenetic analysis was conducted based on the gene encoding the glycoprotein of equine herpesvirus type 1. It was determined (Fig. 2) that the investigated variant of equine herpesvirus forms a cohesive group with viruses from Europe, China and CIS countries.

As a result of molecular genetic analysis of samples from horses exhibiting clinical signs of Equine Rhinopneumonitis, it was demonstrated that the samples contained nucleic acids of equine herpesvirus type 1. The assembly of the complete genome of the virus was conducted, indicating that the investigated virus belongs to a unified group of viruses circulating in Europe, China and the CIS countries.

Table 2: The results of clinical examination and laboratory analyses for three model farms

Characteristic	Mean±SD	Median	Interquartile range	Total number of	Incidence rate
	(proportion)	(proportion)	(proportion)	cases	(%)
Nasal discharge	0.25±0.15	0.20	0.15-0.35	52	26
Cough	0.30±0.20	0.25	0.20-0.40	63	31.5
Fever	38.5±0.3	38.4	38.2–38.7	108	54
Apathy	0.15 ± 0.10	0.12	0.10-0.20	34	17
Serological diagnosis	0.50±0.25	0.45	0.40-0.65	52	26
Virological diagnosis	0.40 ± 0.20	0.35	0.30-0.50	44	22
Molecular-genetic	0.60±0.30	0.55	0.50-0.75	62	31

Note: Clinical symptom data (nasal discharge, cough, apathy) are presented as mean \pm standard deviation (SD). Data on nasal discharge, cough, and apathy are measured in units, where 0 indicates the absence of the symptom, and values greater than 0 assess the severity of the symptom. Data on temperature elevation are presented as the mean value in degrees Celsius. Laboratory analysis data (serological, virological, molecular-genetic diagnostics) are presented as the mean proportion with positive results \pm standard deviation (SD).

Table 3: The anal	vsis of disease	prevalence in	different age groups

Age Group	Mean±SD	Median	Interquartile range	Total number of cases	Incidence rate (%)
Young	0.40 ± 0.20	0.35	0.30-0.50	82	41
Adults	0.45 ± 0.25	0.40	0.35-0.55	95	47.5
Seniors	0.55±0.30	0.50	0.45-0.65	110	55

Table 4: The comparison between 2022 and 2023 and between farms							
Variable	2022 year	2023 year	Farm in Akmola Region	Farm in Pavlodar Region	Farm in North Kazakhstan Region		
The mean number of cases	15±5	20±7	10±3	18±6	25±8		
Total number of cases	46	68	30	56	75		
Incidence rate (%)	23	34	15	28	37.5		

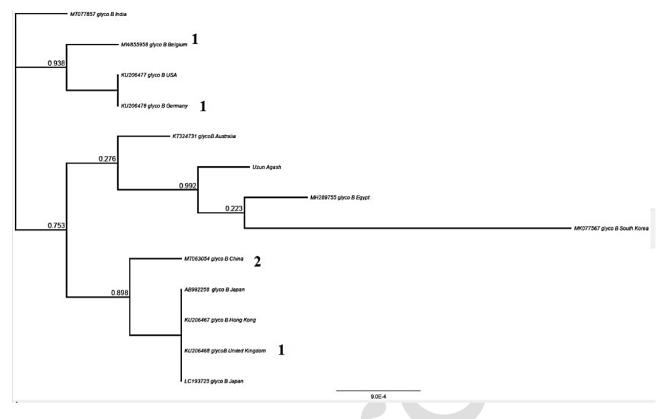


Fig. 2: Phylogenetic analysis of equine herpesvirus variants based on the model of glycoprotein B.

DISCUSSION

The obtained results allow us to conclude that Equine Rhinopneumonitis is a prevalent and potentially dangerous disease for horses in regions where it is registered. Furthermore, the presence of differences in morbidity among different age groups and between farms indicates the importance of an individualized approach to vaccination and disease control depending on the specific conditions of horse management (Pusterla et al. 2022; Kovalcuka et al. 2024).

The high effectiveness of the applied treatment and prevention methods, confirmed by the absence of disease cases in all three farms in 2023, underscores the importance of a systemic approach to managing the epidemiological situation and maintaining the health of animals in equine facilities (Osterrieder et al. 2023; Goehring et al. 2024). Similar findings have been observed in other studies, highlighting the role of vaccination and biosecurity measures in controlling the spread of Equine Rhinopneumonitis (Ferlazzo et al. 2020; Khusro et al. 2020).

The results of genetic research also hold significant value as they help identify the genetic characteristics of the Equine Rhinopneumonitis virus and its relationship with other known strains. This may contribute to the development of more effective diagnostic and vaccination methods in the future (Lupulovic et al. 2021; Ramsauer et al. 2021). Studies have shown that the genetic variability of EHV-1 and EHV-4 plays a crucial role in disease severity and transmission, further emphasizing the need for continued genomic surveillance (El-Hage et al. 2021).

Thus, the findings of this study can serve as a basis for

the development and implementation of control and prevention strategies for Equine Rhinopneumonitis in horses not only in Kazakhstan but also in other countries with similar epidemiological situations (Ventsova and Safonov 2021). Considering global trends, integrating molecular diagnostics with epidemiological surveillance can improve early detection and intervention efforts, ultimately reducing economic losses and improving animal welfare.

The findings of the study by Deniz et al. (2023) revealed that 52.6% of the samples tested positive by Real-Time PCR and 39.4% tested positive by IHC assays. A correlation between these two diagnostic methods was observed. Histopathological analysis revealed inclusions in the lungs, spleen, and predominantly in the liver in 52.6% of the foals. Similarities with our study lie in the utilization of different diagnostic methods to detect the disease and verify their correlation, which is crucial for establishing accurate diagnosis and disease control. According to Studer (2023), older horses and those infected with neurotropic strains have a higher risk of developing EHM. Although only the humoral immune response was monitored in this study, not the cellular immune response, the results confirm that a strong antibody response may reduce viral shedding alleviate the severity and possibly of clinical manifestations but may not provide complete protection against complications. This study resembles ours in that it also analyzes the course of EHV-1 infection in horses and identifies important influencing factors disease development and the effectiveness of the immune response.

The findings of the study by Worku et al. (2024a) are of interest, as they established that donkeys had twice the chance of contracting EHV compared to horses. The prevalence of EHV-1/-4 was higher in horses aged 3 to 8 years and lower in horses younger than 3 years, with mares having 1.73 times higher chances of contracting EHV compared to stallions. A higher prevalence was detected in lactating mares and pregnant horses. Overall, this study indicates a high and widespread prevalence of EHV-1/-4 in the investigated area (Amhara), warranting due attention. Developing strategies for preventing and minimizing the spread and occurrence of infection is extremely important. This study also shares similarities with our data, as both assess the prevalence and epidemiological risk factors of EHV infection in horses. Both studies provide important information about the distribution and spread of this pathogen, which can be useful in developing strategies for controlling and preventing this infection. Study by Tekelioğlu and Ömer (2023) utilized PCR to detect and differentiate between EHV-1 and EHV-4 viruses in horses in Turkey. PCR successfully identified and differentiated both viruses in tissue samples and nasal swabs, confirming the presence of both viruses among the racehorse population in Turkey. This study is similar to ours in using PCR for virus diagnosis and confirming its presence in the horse population. The findings of the study by Worku et al. (2024b) in Ethiopia showed a 65.9% prevalence of equine herpesvirus, with higher rates among older horses and females. Similar to our study, it identifies risk factors and underscores the importance of controlling the transmission of EHV-1/-4. Balena et al. (2023) evaluates genetically modified variants of Equid alphaherpesvirus 1 (EHV-1) to create improved vaccines. Removal of genes associated with virulence and immune evasion led to a significant reduction in virulence in vitro and in vivo. These mutants elicited enhanced immune responses, making them potential vaccine candidates. This is similar to our study, where EHV-1 variants were also evaluated for vaccine development. A study by Bannai et al. (2023) indicates partial protection mediated by systemic antibodies and mucosal IgG responses. It is also known that stress induced by seasonal changes and management practices played a role in the occurrence of disease outbreaks in horses (Pavulraj et al. 2021). The conclusions of the study by Khattab et al. (2022) confirm the involvement of EHV-4 in horse abortions, suggesting the possibility of reactivation of latent virus under the influence of stress and other factors. Finally, according to the findings of the study by Hassanien et al. (2020), the detection of EHV-1 and AHV-5 in aborted fetuses of Arabian horses in Egypt using PCR and virus isolation is confirmed. This is the first detection and isolation of AHV-5 in Egypt, as well as the first case of AHV-1 detection in an aborted fetus worldwide. These results indicate the widespread distribution of infections among horses and underscore the importance of combating these viruses.

Conclusion

The study on the prevalence of Equine Rhinopneumonitis in Kazakhstan allows for several important conclusions.

Firstly, the analysis of epizootiological data indicates the widespread occurrence of the disease across the country. Over the past decade, Equine Rhinopneumonitis has been detected in various regions, with the number of epizootic outbreaks increasing each year. The highest number of outbreaks was recorded in 2021. This highlights the necessity of strengthening measures for disease control and prevention in the country.

Secondly, the results of the analysis of clinical examination data and laboratory tests confirm the significance of laboratory diagnostic methods. Statistical tests have shown statistically significant differences between symptom groups and a positive correlation between nasal discharge and virological diagnosis. This underscores the need for a comprehensive approach to disease diagnosis and monitoring.

Thirdly, the analysis of morbidity in different age groups has shown that the prevalence of rhinopneumonitis may vary depending on the age of the horses. This is an important observation that can be useful in developing prevention and treatment strategies.

Finally, the results of genetic research confirm the presence of equine herpesvirus type 1 and its belonging to a specific group of viruses circulating in Europe, China, and CIS countries. This provides insight into the origin and characteristics of the virus, which can be valuable for the development of vaccines and disease control methods.

In conclusion, the findings of this study provide important information regarding the prevalence and characteristics of Equine Rhinopneumonitis in horses in Kazakhstan, which can be utilized for the development and enhancement of measures to combat this disease. The obtained results have both practical and scientific significance and can be applied not only in Kazakhstan but also in other countries to improve the control and management of Equine Rhinopneumonitis in horses. The practical application of the results includes the development of effective vaccination strategies and disease control, especially in areas with high incidence rates. The scientific significance of the study lies in its contribution to expanding our understanding of the epidemiology, clinical manifestations, and genetic characteristics of the Equine Rhinopneumonitis virus. However, a limitation of this study is that it only encompasses data collected over a specific period and in particular territories of Kazakhstan, which may limit the generalizability of the results.

In the future, one direction for further research could involve a deeper investigation into the molecular mechanisms of interaction between the Equine Rhinopneumonitis virus and the horse's organism, which could aid in the development of more effective diagnostic and treatment methods. Additionally, an important area of research could involve assessing the effectiveness of various vaccination and disease control strategies in practice, as well as their economic efficiency for equine establishments and veterinary services.

Conflict of interest: This research has no conflict of interests.

Funding: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Availability of data and material: Data will be available on request.

Author's Contribution: Conceptualization: AS, SN, KK. Data curation: DK, AB, YM. Formal analysis: MA, AA, BK. Funding acquisition: NS. Methodology: AS, SN, KK. Project administration: NS. Visualization: DK, AB, YM. Writing - original draft: AB, SN, NS, KK. Writing - review & editing: AS, DK.

REFERENCES

- Balena V, Pradhan SS, Bera BC, Anand T, Sansanwal R, Khetmalis R, Madhwal A, Bernela M, Supriya K, Pavulraj S and Tripathi BN, 2023. Double and quadruple deletion mutant of EHV-1 is highly attenuated and induces optimal immune response. Vaccine 41(5): 1081-1093. https://doi.org/10.1016/j.vaccine.2022.12.044
- Bannai H, Kambayashi Y, Nemoto M, Ohta M and Tsujimura K, 2023. Experimental challenge of horses after prime-boost immunization with a modified live equid alphaherpesvirus 1 vaccine administered by two different routes. Archives of Virology 168(1): 27. https://doi.org/10.1007/s00705-022-05638-w
- Broux B, Gryspeerdt A, Amory H, Frippiat T, Gasthuys F, Legrand L and Deprez P, 2016. Prevalence of respiratory pathogens in nasal swabs from horses with acute respiratory disease in Belgium. Vlaams Diergeneeskundig Tijdschrift 85(4): 221-224. https://doi.org/10.21825/vdt.v85i4.16332
- Couroucé A, Normand C, Tessier C, Pomares R, Thévenot J, Marcillaud-Pitel C, Legrand L, Pitel PH, Pronost S and Lupo C, 2023. Equine herpesvirus-1 outbreak during a showjumping competition: A clinical and epidemiological study. Journal of Equine Veterinary Science 128: 104869. https://doi.org/10.1016/j.jevs.2023.104869
- Dayaram A, Seeber PA and Greenwood AD, 2021. Environmental detection and potential transmission of equine herpesviruses. Pathogens 10(4): 423. https://doi.org/10.3390/pathogens10040423
- Deniz S, Gülçubuk A, Sönmez K, Altan E, Danyer IA, Gürel A and Yılmaz H, 2023. Investigation of the presence of equine herpesvirus-1 (EHV-1) in tissue samples of aborted foals by histopathological, immunohistochemical, and Real-Time PCR techniques. Journal of the Hellenic Veterinary Medical Society 74(4): 6695-6706. https://doi.org/10.12681/jhvms.34093
- El Brini Z, Fassi Fihri O, Paillot R, Lotfi C, Amraoui F, El Ouadi H, Dehhaoui M, Colitti B, Alyakine H and Piro M, 2021. Seroprevalence of equine herpesvirus 1 (EHV-1) and equine herpesvirus 4 (EHV-4) in the Northern Moroccan horse populations. Animals 11(10): 2851. https://doi.org/10.3390/ani11102851
- El-Hage C, Mekuria Z, Dynon K, Hartley C, McBride K and Gilkerson J, 2021. Association of Equine Herpesvirus 5 with Mild Respiratory Disease in a Survey of EHV1,-2,-4 and-5 in 407 Australian Horses. Animals 11(12): 3418. <u>https://doi.org/10.3390/ani11123418</u>
- Ferlazzo A, Cravana C, Fazio E and Medica P, 2020. The different hormonal system during exercise stress coping in horses. Veterinary World 13(5): 847-859. https://doi.org/10.14202%2Fvetworld.2020.847-859
- Goehring L, Dorman DC, Osterrieder K, Burgess BA, Dougherty K, Gross P, Neinast C, Pusterla N, Soboll-Hussey G and Lunn DP, 2024. Pharmacologic interventions for the treatment of equine herpesvirus-1 in domesticated horses: A systematic review. Journal of Veterinary Internal Medicine 38(3): 1892-1905. <u>https://doi.org/10.1111/jvim.17016</u>
- Gomez DE, Arroyo LG, Lillie B and Weese JS, 2021. Nasal bacterial microbiota during an outbreak of equine herpesvirus 1 at a farm in southern Ontario. Canadian Journal of Veterinary Research 85(1): 3-11.
- Hassanien RT, El-Nahas EM, Mahmoud NA and El-Bagoury GF, 2020. Molecular characterization of equine herpesvirus-1 and asinine herpesvirus-5 isolated from aborted fetuses of

Arabian horses. The Thai Journal of Veterinary Medicine 50(3): 397-403. <u>https://doi.org/10.56808/2985-1130.3042</u>

- Khattab OM, Abdelmegeed HK, Mashaly MM, Hamdy M, Hagag N, Hamed A, Fahmy HA, Ibrahim E, Shahein MA and Ahmed EM, 2022. Equine herpes virus 4 (EHV4) investigation in aborted Egyptian Mares; molecular detection, isolation, and phylogeny for viral glycoprotein B. Advances in Animal and Veterinary Sciences 10(9): 1907-1915. https://doi.org/10.17582/journal.aavs/2022/10.9.1907.1915
- Khusro A, Aarti C, Rivas-Caceres RR and Barbabosa-Pliego A, 2020. Equine herpesvirus-I infection in horses: Recent updates on its pathogenicity, vaccination, and preventive management strategies. Journal of Equine Veterinary Science 87: 102923. https://doi.org/10.1016/j.jevs.2020.102923
- Kovalcuka L, Mālniece A and Vanaga J, 2024. Comparison of Tonovet® and Tonovet plus® tonometers for measuring intraocular pressure in dogs, cats, horses, cattle, and sheep. Veterinary World 17(2): 384-388.
 <u>http://doi.org/10.14202/vetworld.2024.384-388</u>
- Krisová Š, Tóthová K, Molinková D, Makra Z and Zisopoulou AM, 2020. Prevalence of equine herpesvirus 2 (EHV-2) in equine ocular disease. Acta Veterinaria Brno 89(2): 115-123. https://doi.org/10.2754/avb202089020115
- Laval K, Poelaert KC, Van Cleemput J, Zhao J, Vandekerckhove AP, Gryspeerdt AC, Garré B, van der Meulen K, Baghi HB, Dubale HN and Zarak I, 2021. The pathogenesis and immune evasive mechanisms of equine herpesvirus type 1. Frontiers in Microbiology 12: 662686. https://doi.org/10.3389/fmicb.2021.662686
- Lupulovic D, Savić S, Gaudaire D, Berthet N, Grgić Ž, Matović K, Deshiere A and Hans A, 2021. Identification and genetic characterization of equine infectious anemia virus in Western Balkans. BMC Veterinary Research 17: 168. https://doi.org/10.1186/s12917-021-02849-2
- Mahmoud HY, Fouad SS and Amin YA, 2023. Review of two viral agents of economic importance to the equine industry (equine herpesvirus-1, and equine arteritis virus). Equine Veterinary Education 35(2): 92-102. https://doi.org/10.1111/eve.13649
- Marenzoni ML, De Waure C and Timoney PJ, 2023. Efficacy of vaccination against equine herpesvirus type 1 (EHV-1) infection: systematic review and meta-analysis of randomised controlled challenge trials. Equine Veterinary Journal 55(3): 389-404. <u>https://doi.org/10.1111/evj.13870</u>
- Osterrieder K, Dorman DC, Burgess BA, Goehring LS, Gross P, Neinast C, Pusterla N, Hussey GS and Lunn DP, 2023. Vaccination for the prevention of equine herpesvirus-1 disease in domesticated horses: A systematic review and meta-analysis. Journal of Veterinary Internal Medicine 38(3): 1858-1871. https://doi.org/10.1111/jvim.16895
- Parameswaran T and Senthil NR, 2014. Analysis of economic loss due to equine herpes viral infection. International Journal of Advanced Veterinary Science and Technology 3(1): 134-139. <u>https://doi.org/10.23953/cloud.ijavst.196</u>
- Pavulraj S, Eschke K, Theisen J, Westhoff S, Reimers G, Andreotti S, Osterrieder N and Azab W, 2021. Equine herpesvirus type 4 (EHV-4) outbreak in Germany: Virological, serological, and molecular investigations. Pathogens 10(7): 810. https://doi.org/10.3390/pathogens10070810
- Pusterla N, Hatch K, Crossley B, Wademan C, Barnum S and Flynn K, 2020. Equine herpesvirus-1 genotype did not significantly affect clinical signs and disease outcome in 65 horses diagnosed with equine herpesvirus-1 myeloencephalopathy. The Veterinary Journal 255: 105407. <u>https://doi.org/10.1016/j.tvjl.2019.105407</u>
- Pusterla N, James K, Barnum S, Bain F, Barnett DC, Chappell D, Gaughan E, Craig B, Schneider C and Vaala W, 2022. Frequency of detection and prevalence factors associated

with common respiratory pathogens in equids with acute onset of fever and/or respiratory signs (2008-2021). Pathogens 11(7): 759. https://doi.org/10.3390/pathogens11070759

- Pusterla N, Mapes S and David Wilson W, 2012. Prevalence of latent alpha-herpesviruses in thoroughbred racing horses. The Veterinary Journal 193(2): 579-582. <u>https://doi.org/10.1016/j.tvjl.2012.01.030</u>
- Ramsauer AS, Badenhorst M and Cavalleri JM, 2021. Equine parvovirus hepatitis. Equine Veterinary Journal 53(5): 886-894. <u>https://doi.org/10.1111/evj.13477</u>
- Safonov VA, Mikhalev VI and Chernitskiy AE, 2018. Antioxidant status and functional condition of respiratory system of newborn calves with intrauterine growth retardation. Agricultural Biology 53(4): 831-841. https://doi.org/10.15389/agrobiology.2018.4.831eng
- Studer JL, 2023. Longitudinal study on the virus-host interaction in acutely EHV-1 infected horses. Doctoral dissertation. Zurich: University of Zurich.
- Tekelioğlu BK and Ömer AK, 2023. Detecting Equine Herpesvirus-1 and Equine Herpesvirus-4 Infections in Racehorses using Polymerase Chain Reaction. Black Sea Journal of Health Science 6(4): 705-712. <u>https://doi.org/10.19127/bshealthscience.1349444</u>

- Ventsova I and Safonov V, 2021. The role of oxidative stress during pregnancy on obstetric pathology development in high-yielding dairy cows. American Journal of Animal and Veterinary Sciences 16(1): 7-14. https://doi.org/10.3844/ajavsp.2021.7.14
- Worku A, Molla W, Kenubih A, Gizaw D, Muluneh A, Admassu B, Ejo M, Dagnaw GG, Bitew AB, Fentahun T and Getnet K, 2024a. Seroprevalence and associated risk factors of equine herpesvirus type-1/-4 in selected districts of Northwest Amhara, Ethiopia. Comparative Immunology, Microbiology & Infectious Diseases 107: 102155. <u>https://doi.org/10.1016/j.cimid.2024.102155</u>
- Worku A, Molla W, Kenubih A, Gizaw D, Muluneh A, Admassu B, Ejo M, Dagnaw GG, Bitew AB, Fentahun T and Getnet K, 2024b. Comparative immunology, microbiology and infectious diseases. Comparative Immunology, Microbiology & Infectious Diseases 107: 102155. https://doi.org/10.1016/j.cimid.2024.102155
- Zarski LM, Giessler KS, Jacob SI, Weber PS, McCauley AG, Lee Y and Soboll Hussey G, 2021. Identification of host factors associated with the development of equine herpesvirus myeloencephalopathy by transcriptomic analysis of peripheral blood mononuclear cells from horses. Viruses 13(3): 356. https://doi.org/10.3390/v13030356