

https://www.ufpr.br/

# Seroepidemiological study of Crimean-Congo hemorrhagic fever (CCHF) in Wasit governorate, Iraq 2024: a preliminary assessment

Submitted: 02/02/2025 Accepted: 05/04/2025

Thulfigar Husain Ali Abu-al Leil<sup>1\*</sup>, Israa Abdulwadood Muhammed Ali Alsaad<sup>2</sup>

Correspondence email: <u>faqarhusain@gmail.com</u>

Abstract: Crimean-Congo hemorrhagic fever (CCHF) is a serious disease that leads to severe symptoms in humans and animals. The fact that Crimean-Congo hemorrhagic fever is asymptomatic in animals poses a danger. In the last few years, there has been an increase in the incidence of CCHF in humans, with a high mortality rate in Iraq. The current study was conducted to address CCHF in cattle in the Wasit governorate, Central Iraq. The study includes an investigation into the presence of antibodies for the CCHFV in sheep and goats using the CCHF Double Antigen Multi-Species-CCHFDA ELISA kit to determine the existence of IgG antibodies against the CCHFV, as well as their correlation with the species, age, and sex of the animals. One hundred ninety-six serum samples were collected from private farms and distributed in different Wasit Governorate areas, including 104 sheep and 92 goats. The results showed an overall seroprevalence rate of 35.6% in sheep and 39.1% in goats. This indicates a severe warning and a likelihood of a rapid increase to spread to other areas, which directly impacts public health. Therefore, we recommend conducting further studies to investigate this disease more comprehensively. This will enhance our understanding of its nature and aid in the development of specialized programs for tick control. These would include the use of repellents and acaricides, implementation of environmental management systems, application of tick control measures in animal pens, adoption of biological control methods, and organizing awareness. Training programs for breeders are essential to enhance their knowledge and practices in tick prevention and management to limit and control its spread.

Keywords: Tick born disease; Small ruminant; Nairoviridae; ELISA; Zoonosis.

#### 1. Introduction

Crimean Congo hemorrhagic fever (CCHF) is a highly infectious illness caused by the virus of the family Nairoviridae, genus Orthonairovirus, which lacks any approved treatments or vaccines, with supportive treatment being the first form of treatment and initially discovered in 1944 in the Peninsula of Crimean following an outbreak among military members, leading to the disease being referred to as Crimean hemorrhagic fever (Hoogstraal, 1979; Whitehouse, 2004). Infection with CCHFV primarily occurs by being bitten by a tick or by contact directly with blood and/or tissue from a diseased animal, transmission from person to person, especially in hospitals (Faraj et al., 2021; Pshenichnaya and Nenadskaya, 2015; Tsergouli et al., 2020). Around 10,000 to 15,000 cases of CCHF are believed to spread globally every year, but getting exact figures is challenging. This is because as many as 88% of these cases might not show any symptoms (Ergonul and Whitehouse, 2007; Mishra et al., 2022).

The broad spread across different areas on the planet, along with its ability to cause deadly illnesses in humans, is currently a significant issue for public health (Saleh et al., 2025). Additionally, the disease is expected to move into new areas as the world becomes more interconnected and the climate and environment change (Leblebicioglu, 2010; Purnak et al., 2007). Viruses may infect various human cells, causing direct harm due to the viral infection and indirectly by altering the permeability of blood vessels and triggering a pro-inflammatory immune reaction (Akıncı et al., 2013). CCHFV-related illness is restricted to humans, but it has been observed that asymptomatic short-term viral infections can occur in various animals, including livestock and wild animals, with viremia lasting for as long as 15 days (Nurettin et al., 2022; Hashim and Al-Hamed, 2024).

Iraq, a country in the eastern Mediterranean, documented the first case of CCHF in 1979, the initial diagnosis in ten individuals (Al-Tikriti et al., 1981). Over the years, the number of reported cases has fluctuated, with a peak of six from 1989 to 2009, followed by a significant increase to 11 cases in 2010 (Majeed et al., 2012; S Al-Yabis et al., 2005). The year 2018 saw a particularly tragic turn of events, with three deaths reported from the disease from 10 cases positively confirmed with PCR(WHO, 2022; Dakhil et al., 2024). By 2021, a total of 33 confirmed cases, including 13 fatalities. In 2022, 97 suspected cases were verified via (PCR) Polymerase Chain Reaction testing conducted by the Iraq Central Public Health Laboratory, where 13(13%) deaths were directly linked to the disease (Alhilfi et al., 2023a; Jafar et al., 2022). In 2023, 511 cases of confirmed CCHF infection were reported through PCR testing (Atwan et al., 2024). The objective of the present study is to determine the seroprevalence of CCHF infection in sheep and goats in the Wasit government in Iraq.

## 2. Materials and Methods

## 2.1. Animals and area of study

The current study included 196 animals, comprising 104 sheep and 92 goats, randomly selected from both rural and urban areas of Wasit Governorate. The sampled areas included Al-Suwaira, Al-Aziziya, Al-Nu'maniya, Al-Kut, Al-Mufaqiya, Badra, Al-Hai, and Sheikh Saad. At sampling, all animals were infested with ticks, and the data included species, age, gender, type of ownership, and location.

https://doi.org/10.5380/avs.v30i2.98304



<sup>&</sup>lt;sup>1</sup>Animal Wealth Department, Wasit Agriculture Directorate, Ministry of Agriculture, Wasit, Iraq, E-mail: <u>faqarhusain@gmail.com</u>, ORCID https://orcid.org/0009-0007-8968-3977

<sup>&</sup>lt;sup>2</sup>Department of Internal and Preventive Veterinary Medicine, College of Veterinary Medicine, University of Basrah, Basra / Iraq, E-mail: IsraAli@uobasrah.edu.iq



### 2.2. Samples collection

Blood was taken in accordance to Jackson (2013). Five milliliters of blood were taken from the jugular vein aseptically and put in a tube to separate the serum. The gel tube was stored in a cool box and transported to the lab, where the serum was separated by centrifuge and stored in deep freeze until the day of analysis.

### 2.3. Ethical approval

The samples from sheep and goats in this study were collected according to all applicable international and national regulations and guidelines, as well as the ethical and humane principles followed by Veterinary Medicine College, Basrah University, Iraq, and ensuring that the animals from which the samples were taken were not subjected to stress or pain.

### 2.4. Serological analysis

The sera of animals were analyzed for anti-CCHFV IgG antibodies utilizing the "ID Screen CCHF Double Antigen Multi-species – CCHFDA" ELISA kit from (ID. Vet, Grabels, France). Serological analysis was accomplished by following the directions provided by the manufacturer. Each microplate was evaluated at an optical density of 450 nm utilizing the HumaReader HS ELISA microplate reader (Human, Wiesbaden, Germany). Samples presenting (SP) were counted by the formula provided by the manufacturer classifying samples as negative and positive:

SP(%) = (mean of sample's optical density /mean of positive's control optical density)  $\times 100$ 

Results with SP more than 30% were deemed +ve, while those with SP less than 30% were considered -ve.

#### 2.5. Statistical analysis:

The data was evaluated using biostatistics, with significance set at P<0.05, employing IBM SPSS Statistics version 20 (New York, United States). Chi-square tests were performed to determine the association between factors and infections.

#### 3. Results

The antibodies titer of CCHFV in sheep and goat sera evaluated using the "ID Screen CCHF Double Antigen Multi-species-CCHFDA ELISA Kit" in Wasit was 73 (37.2%) out of 196 serum samples, comprising 37 sheep (35.6%) and 36 goats (39.1%) with no significant difference ( $p \ge 0.05$ ) (Table 1). The current study found that sheep and goats older than three years had the highest seroprevalence (52.1%), followed by animals between two and three years (24.7%) (Table 2). There was a considerable variation in the seroprevalence of CCHFV between male and female animals ( $p \le 0.05$ ), as seen in Table 3.

	CCHF -	CCHF +	Total
sheep	(67) 64.4%	(37) 35.6%	104
goat	(56) 60.9%	(36) 39.1%	92
Total	(123) 62.8%	(73) 37.2%	196

Table 1 – Seroprevalence of IgG antibodies against CCHFV separated by species.

	CCHF -	CCHF +	Total
less than 1 year	17(13.8%)	12(16.4%)	29(14.8%)
1 - 2 year	20(16.3%)	5(6.8%)	25(12.8%)
2 - 3 year	41(33.3%)	18(24.7%)	59(30.1%)
More than 3 years	45(36.6%)	38(52.1%)	83(42.3%)

Table 2 – Seroprevalence of the IgG antibody against CCHFV separated age categories.

	CCHF -	CCHF +	Total
FEMALE	110(66.3%)	56(33.7%)	166
MALE	13(43.3)	17(56.7%)	30

Table 3 – Seroprevalence of the IgG antibody against CCHFV separated by gender categories.

https://doi.org/10.5380/avs.v30i2.98304



https://revistas.ufpr.br/veterinary





Authors should exclusively cover the results of the experiment, without references. Symbols and units should be represented by: 36.4%, 88.4 kg, 42 ml, (P = 0.045), R2 = 0.89. Always include the mean and standard deviation values (example) in the tables and graphs. Authors should not repeat data between the text and the tables and graphs. Table and chart titles should be self-explanatory.

#### 4. Discussion

In the current study, the seroprevalence rate of CCHF antibodies in serum samples was 37.2% from 196 overall serum samples, including sheep at 35.6% and goats at 39.1%, as determined by using ELISA. This investigation is the first assessment of the seroepidemiological condition of small ruminants (sheep and goats) within the Wasit governorate of Iraq. However, scant information exists regarding the frequency and distribution of CCHFV in Iraq.

The present study reveals significant seroprevalence rates in animals. The possibility of cross-border transmission via livestock trade with endemic regions such as Iran and Turkey, and rising temperatures in the territory further complicate the problem (Altaliby et al., 2021; Ergönül, 2006; Mostafavi et al., 2014), Additionally, inadequate public health strategies, substandard medical and veterinary care, and closeness between flocks, butchers, and animals are significant causes of disease transmission between animals and humans in Iraq in recent years. The current study found that CCHF seroprevalence was statistically significant between males and females which was recorded ( $p \le 0.05$ ), showing a higher seroprevalence rate in males compared to females.

The rise in (CCHF) seroprevalence in Iraq can be related to the persistent increase in global temperatures and the extension of the summer season, during which tick activity intensifies. Moreover, illegal trade with neighboring countries endemic to the disease contributes significantly to the disease (AL-Shauwreed et al., 2023; Alhilfi et al., 2023b; Maltezou and Papa, 2010). In 1996, veterinary authorities established a program to tackle tick infestations in Iraq, reducing yearly CCHF cases (S Al-Yabis et al., 2005). From 1998 to 2009, the number of instances fluctuated between 0 and 6; however, in 2010, there was a significant rise to 11 cases, attributed to the termination of the program and the absence of tick control measures (Majeed et al., 2012). This confirms the necessity of doing laboratory testing routinely to identify and examine illnesses in cattle and assess the severity and spread of ticks (Alhilfi et al., 2023a). Implementing urgent protective control measures is crucial, beginning with selecting relevant tick medicines and assuring their quality and efficacy. Furthermore, initiatives for spraying barns and cattle and dipping small ruminants should be implemented (Ghiasi et al., 2018; Levin, 2020; Maltezou and Papa, 2010).

#### 5. Conclusion

This study shows that small ruminants in the Wasit department in central Iraq have high CCHFV-specific IgG antibodies. This indicates that these animals are good as sentinel animals for CCHFV seroepidemiological monitoring to determine whether CCHFV is present in this area. Nonetheless, CCHF poses a risk to human and public health in Iraq, especially in Wasit, which has many domestic animals, and the nearby governorates depend on Wasit as a food source. The significant seroprevalence rate in goats and sheep, and insufficient tick eradication efforts contribute to the problem.

**Acknowledgments**: The authors would like to thank everyone who helped complete this work, and the owners who consented to use their animals in this study. We also thank the Central Veterinary Laboratories/Iraq staff, particularly the manager and the Virology and Parasitology Department, for their support.

Conflict of Interest: The authors declare no conflicts of interest.

#### 6. References

- AK1NC1, E., BODUR, H. & LEBLEBICIOGLU, H. 2013. Pathogenesis of Crimean-Congo hemorrhagic fever. *Vector-Borne and Zoonotic Diseases*, 13, 429-437.
- AL-SHAUWREED, A. K. M., HAMADI, S. S., ISSA, A. H., SAUD, H. A., ALSHAMI, I. J. J. & FARES, M. N. 2023. Evolutionary and Historical Study of Crimean-Congo Hemorrhagic Fever Virus (CCHFV). *Medical Journal of Basrah University*, 41, 12-25.
- AL-TIKRITI, S., AL-ANI, F., JURJI, F., TANTAWI, H., AL-MOSLIH, M., AL-JANABI, N., MAHMUD, M., AL-BANA, A., HABIB, H. & AL-MUNTHRI, H. 1981. Congo/Crimean haemorrhagic fever in Iraq. *Bulletin of the World Health Organization*, 59, 85.
- ALHILFI, R. A., KHALEEL, H. A., RAHEEM, B. M., MAHDI, S. G., TABCHE, C. & RAWAF, S. 2023a. Large outbreak of Crimean-Congo haemorrhagic fever in Iraq, 2022. *IJID Regions*, 6, 76-79.
- ALHILFI, R. A., KHALEEL, H. A., RAHEEM, B. M., MAHDI, S. G., TABCHE, C. & RAWAF, S. 2023b. Large

- outbreak of Crimean-Congo haemorrhagic fever in Iraq, 2022. *IJID Reg*, 6, 76-79.
- ALTALIBY, M., ESMAEEL, S. & HUSSAIN, K. J. 2021. Seroprevalence of Crimean-Congo Haemorrhagic Fever in sheep and goats in Iraq. *Bulg. J. Vet. Med*, 26, 1-6.
- ATWAN, Z., ALHILFI, R., MOUSA, A. K., RAWAF, S., TORRE, J. D. L., HASHIM, A. R., SHARQUIE, I. K., KHALEEL, H. & TABCHE, C. 2024. Alarming update on incidence of Crimean-Congo hemorrhagic fever in Iraq in 2023. *IJID Regions*, 10, 75-79.
- DAKHIL, A. A., SALEH, W. M. M., AL-TAHER, S. S. H., ABBAS, K., ALQAISI, K., NAJI, M. ABDULRASOOL, M. 2024. L. SEROEPIDEMIOLOGICAL SURVEY OF CRIMEAN-CONGO HEMORRHAGIC FEVER (CCHF) IN SMALL **RUMINANTS** AFTER Α RECENT HUMAN GOVERNORATE, **OUTBREAK** IN **BASRA** SOUTHERN IRAQ IN 2023.

https://doi.org/10.5380/avs.v30i2.98304







- ERGÖNÜL, Ö. 2006. Crimean-Congo haemorrhagic fever. *The Lancet infectious diseases*, **6**, 203-214.
- ERGONUL, O. & WHITEHOUSE, C. A. 2007. *Crimean-Congo hemorrhagic fever: a global perspective*, Springer Science & Business Media.
- FARAJ, K. B., SAUD, S. S. & ALMYAHII, M. H. 2021. Study of the Common Tick Species of Cattle in the North of Basrah Province. *Annals of the Romanian Society for Cell Biology*, 25, 6740-6748.
- GHIASI, H., LUPI, T. M. & MOKHTARI, M. S. 2018. The estimation of genetic parameters for growth curve traits in Raeini Cashmere goat described by Gompertz model. *Small Ruminant Research*, 165, 66-70.
- HASHIM, D. A. & AL-HAMED, T. A. 2024. Seroprevalence of Crimean-Congo Hemorrhagic Fever in Cattle in Basrah Province, Iraq. *Basrah Journal of Veterinary Research*, 23, 118-129.
- HOOGSTRAAL, H. 1979. Review Article1: The Epidemiology of Tick-Borne Crimean-Congo Hemorrhagic Fever in Asia, Europe, and Africa23. *Journal of Medical Entomology*, 15, 307-417.
- JACKSON, M. L. 2013. *Veterinary clinical pathology: an introduction*, John Wiley & Sons.
- JAFAR, U., USMAN, M., EHSAN, M., NAVEED, A., AYYAN, M. & CHEEMA, H. A. 2022. The outbreak of Crimean-Congo hemorrhagic fever in Iraq - Challenges and way forward. *Annals of Medicine and Surgery*, 81, 104382.
- LEBLEBICIOGLU, H. 2010. Crimean-Congo haemorrhagic fever in Eurasia. *Int J Antimicrob Agents*, 36 Suppl 1, S43-6.
- LEVIN, M. L. 2020. *Tick Control* [Online]. MSD Veterinary Manual: MSD Veterinary Manual. Available: <a href="https://www.msdvetmanual.com/integumentary-system/ticks/tick-control">https://www.msdvetmanual.com/integumentary-system/ticks/tick-control</a> [Accessed 2020].
- MAJEED, B., DICKER, R., NAWAR, A., BADRI, S., NOAH, A. & MUSLEM, H. 2012. Morbidity and mortality of Crimean-Congo hemorrhagic fever in Iraq: cases reported to the National Surveillance System, 1990– 2010. Transactions of The Royal Society of Tropical Medicine and Hygiene, 106, 480-483.
- MALTEZOU, H. C. & PAPA, A. 2010. Crimean—Congo hemorrhagic fever: risk for emergence of new endemic foci in Europe? *Travel medicine and infectious disease*, 8, 139-143.
- MISHRA, A. K., HELLERT, J., FREITAS, N., GUARDADO-CALVO, P., HAOUZ, A., FELS, J. M., MAURER, D. P., ABELSON, D. M., BORNHOLDT, Z. A. & WALKER, L. M. 2022. Structural basis of synergistic neutralization of Crimean-Congo hemorrhagic fever virus by human antibodies. *Science*, 375, 104-109.
- MOSTAFAVI, E., HAGHDOOST, A. A., DOOSTI, I. A., BOKAEI, S. & CHINIKAR, S. 2014. Temporal modeling of Crimean-Congo hemorrhagic fever in Iran.
- NURETTIN, C., ENGIN, B., SUKRU, T., MUNIR, A., ZATI, V. & AYKUT, O. 2022. The seroprevalence of Crimean-Congo Hemorrhagic Fever in wild and domestic animals: an epidemiological update for domestic animals and first

- seroevidence in wild animals from Turkiye. *Veterinary Sciences*, 9, 462.
- PSHENICHNAYA, N. Y. & NENADSKAYA, S. A. 2015. Probable Crimean-Congo hemorrhagic fever virus transmission occurred after aerosol-generating medical procedures in Russia: nosocomial cluster. *International Journal of Infectious Diseases*, 33, 120-122.
- PURNAK, T., SELVI, N. A. & ALTUNDAG, K. 2007. Global warming may increase the incidence and geographic range of Crimean-Congo hemorrhagic fever. *Medical hypotheses*, 68, 924-925.
- S AL-YABIS, A., AL-THAMERY, A. K. & J HASONY, H. 2005. Seroepidemiology of Crimean-Congo haemorrhagic fever in rural community of Basrah. *The Medical Journal of Basrah University*, 23, 30-35.
- SALEH, W., DAKHIL, A., AL-TAHER, S., NAJI, M., ABDULRASOOL, L. & ALQAISI, K. 2025. Seroepidemiological study of crimean-congo hemorrhagic fever (CCHF) in small ruminants in Thi-qar governorate, southern Iraq, 2023. *Adv. Anim. Vet. Sci*, 13, 365-371.
- TSERGOULI, K., KARAMPATAKIS, T., HAIDICH, A., METALLIDIS, S. & PAPA, A. 2020. Nosocomial infections caused by Crimean—Congo haemorrhagic fever virus. *Journal of Hospital Infection*, 105, 43-52.
- WHITEHOUSE, C. A. 2004. Crimean—Congo hemorrhagic fever. *Antiviral Research*, 64, 145-160.
- WHO. 2022. Crimean-Congo hemorrhagic fever Iraq [Online]. World Health Organization. Available: <a href="https://www.who.int/emergencies/disease-outbreak-news/item/2022-DON386">https://www.who.int/emergencies/disease-outbreak-news/item/2022-DON386</a> [Accessed 1 June 2022].

