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Case report

Mers Coronavirus in Dromedary Herd in South Morocco

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Background: Middle East Respiratory Syndrome (MERS) is a zoonotic viral disease that can be transmitted from dromedaries to human beings. Up to December 2016, a total of 1841 cases of laboratory-confirmed MERS-CoV infection were reported to the World Health Organization, with an overall case fatality rate of 35% [1].

The current status of knowledge around MERS-CoV in dromedaries in Morocco still unknow.

This article is a prospective study in dromedary herd during one year 2016 in the south of morocco. Faecal samples and nasal swabs of camels were analyzed for identification of viral sequences using specific polymerase chain reaction (PCR) methods.

Introduction:

Middle East respiratory syndrome coronavirus (MERS-CoV) was first detected in 2012, approximately 1,000 human infections have been reported to the World Health Organization, all linked to residence in or travel to countries on the Arabian Peninsula [2]. Dromedaries (Camelus dromedarius) are thought to play a central role in MERS epidemiology because widespread evidence of MERS-CoV-specific antibodies and virus shedding in camels was found [3], and highly similar viruses have been detected in humans and dromedaries at the same location [4,5].

The Middle East Respiratory Syndrome (MERS-Cov) is considered by the World Health Organization (WHO) to be a serious threat to public health, since it causes severe acute respiratory distress, often-fatal [6]. Infection is widespread in dromedaries, and may be responsible for transmission to humans [7,8,9].

The objective of the study is to detect MERS-Cov in dromedary herds in southern Morocco and to establish an intervention plan to prevent the occurrence of human cases.

Materials and Methods

In collaboration with the National Food Safety Authority (ONSSA) - Marrakech, we collected samples of the upper respiratory tract of 100 dromedaries by swab and stool samples.

Type of study

This is a prospective study, describing and analylizing the MERS-CoV genome in dromedaries in southern Morocco using the RT-PCR research. This study is conducted in the Microbiology-Virology and Biosafety level 2 department of Avicenne Military Hospital of Marrakech (HMA) over a period of 6 months from June 1st to December 31st, 2015.

Study population

The study population is composed of 100 dromedaries from southern Morocco

ICV 2015: 52.82

Inclusion criteria

We included male and female dromedaries of different ages (2 weeks - 17 years), some of which were symptomatic and had a runny nose.

Exclusion criteria

We have excluded dromedaries whose geographical origin is not known.

Results

We divided the dromedaries tested into two age groups: 14 camels (<2 years / 14%) and 86 adults: (>2 years / 86%). The camelin herd contained 19 males (19%) and 81 females (81%) with a sex ratio M/F of 0.23. (**Table 1**)

Ser	Number	Average of age	Extremes
Males	19	3.64	From 2 weeks to 17 years
Females	81	72	From 2 weeks to 16 years
Total	100	6.59	From 2 weeks to 17 years

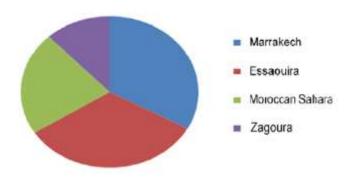
Table 1: Distribution of dromedaries tested by age and sex

19% of the dromedaries tested were symptomatic and showed nasal discharge, they were all adults.

The geographical origin of these dromedaries has shown that 33% of them are from the Essaouira region, 33% from Marrakech, 22% from the Moroccan South and 12% from Zagoura. Dromedaries from these regions may all originate from several regions of the south, as 92% of the camelin herd is located in southern Morocco. (**Figure 1**)

Figure 1: Geographical distribution of dromedaries.

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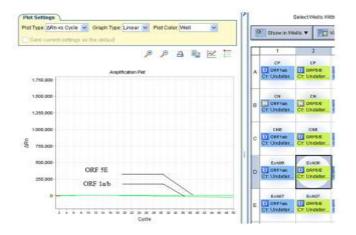


None of the 100 dromedaries analyzed by RT-PCR did not reveal the presence of MERS-Cov, whether for nasal swabs or for stool specimens. (Figure 2, 3)

Figure 2: The results of amplification by RT-PCR of the positive control and the negative control of the ORF1 a / b.



Figure 3: The result of RT-PCR amplification of sample 6, as an example, for ORF 5E and ORF 1a/b



Discussion

MERS-CoV infection is usually confirmed by real-time polymerase chain reaction assays targeting upstream of the E protein gene (upE), open reading frame 1A and open reading frame 1B. Real-time polymerase chain reaction is more sensitive when samples from the lower respiratory tract rather than the upper respiratory tract are tested. However, it is very difficult de make it in dromedaries [2].

Since September 2012, WHO has been notified of 1,952 laboratory-confirmed cases of infection with MERS-CoV and

693 death.

MERS Co-V is a novel coronavirus that belongs to lineage C of the genus betacoronaviruses. It expresses ten open reading frames that encode the production of non-structural proteins as well as structural proteins such as spike (S), envelope (E), nucleocapsid (N) and membrane (M) protein [10]. To gain access to the host cells, MERS-CoV attaches to its functional receptor, human dipeptidyl-peptidase 4 (hDDP4). The receptor is expressed widely in endothelial and subendothelial tissues of multiple organs including the lungs, gastrointestinal tract and kidneys, an observation that may explain the multi-system nature of the associated clinical.

Faecal samples and nasal swabs of camels were analyzed for identification of viral sequences using a specific polymerase chain reaction (PCR) methods. The outcome of this research was negative WHO and the CDC considered dromedaries as the natural reservoir of MERS-CoV [9]. Several studies on dromedary series have been conducted in different countries

Table 2: Percentage of dromedaries positive for the presence of Mers Cov antibodies

Origins	% of dromedaries positive for the presence of MERS CoV antibodies
Oman [11].	100%
Saudi Arabia [12].	90%
Jordan [13].	100%
Ethiopia [14].	97%
Tunisia [14].	54%
Spain [13,14].	14%

The absence of MERS-CoV in Moroccan dromedaries is entirely consistent with the epidemiological situation of the country, which until now has not reported any human cases of MERS-CoV. Moreover, the results of our study do not corroborate with a Spanish study, which suspected 3 dromedaries imported from Morocco 18 years ago, to be the source of infection of the series studied in the Canary Islands. In this respect, it is necessary to conduct a serological study of Moroccan dromedaries in order to confirm or disprove these findings.

In fact, MERS-CoV is widespread in dromedaries in the Middle East, in certain African countries and in the Mediterranean neighbors. Most of these countries have already reported human cases.

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Moreover, this study should encourage the various Moroccan actors, notably the Ministry of Public Health and the Ministry of Agriculture to take the necessary measures to coordinate their monitoring and surveillance systems through regular programs and standardization.

Finally, studies on larger numbers of dromedaries should be considered at national level to make our preliminary work more meaningful.

Conclusion

More than 1800 MERS-CoV infections have been reported so far, with an overall mortality of 35%. The majority of cases are epidemiologically linked to the Middle East. Dromedary camels are the main reservoir for human infection. Although the majority of human cases of MERS have been attributed to human-to-human infections, camels are likely to be a major reservoir host for MERS-CoV and an animal source of MERS infection in humans. However, the exact role of camels in transmission of the virus and the exact route(s) of transmission are unknown.

This preliminary study made it possible to remove the suspicion of carrying MERS-Cov by dromedaries of Moroccan origins. A study on larger numbers is to be envisaged in collaboration with the veterinary services and the relevant departments of the Ministry of Public Health.

References

- 1. World Health Organization (2016) WHO MERS-CoV Global Summary and risk assessment 5 December 2016. http://www.who.int/emergencies/mers-cov/mers-summary-eng.pdf?ua=1 (accessed 7 December 2016)
- 2. World Health Organization Fact sheet Updated May 2017. http://www.who.int/mediacentre/factsheets/mers-cov/en/
- 3. Mackay IM, Arden KE. Middle East respiratory syndrome: an emerging coronavirus infection tracked by the crowd. Virus Res. 2015 [cited 2015 Mar 18]. Epub 2015 Feb 2.
- 4. Haagmans BL, Al Dhahiry SH, Reusken CB, Raj VS, Galiano M, Myers R, et al. Middle East respiratory syndrome coronavirus in dromedary camels: an outbreak investigation. Lancet Infect Dis. 2014;14:140–5. http://dx.doi.org/10.1016/S1473-3099(13)70690-X
- 5. Memish Z, Cotton M, Meyer B, Watson S, Alsahafi A, Al Rabeeah A, et al. Human infection with MERS coronavirus after exposure to infected camels, Saudi Arabia, 2013. Emerg Infect Dis. 2014;20:1012–5.
- 6. Zaki, A. M., S. van Boheemen, T. M. Bestebroer, A. D. Osterhaus, and R. A. Fouchier, 2012: Isolation of a novel coronavirus from a man with pneumonia in Saudi Arabia. N. Engl.J. Med. 367, 1814–1820.
- 7. World organization health for Animals (OIE). Available from: http://www.oie.int/fr/pour-les-medias/communiques-depresse/detail/article/update-august-2014-questions-answers-on-middleeast-respiratory-syndrome-coronavirus-mers-cov/.

- 8. Dépistage en laboratoire du coronavirus du syndrome respiratoire du Moyen-Orient (MERS-CoV) Lignes directrices provisoires Mis à jour juin 2015 WHO/MERS/LAB/15.1 Available from: http://apps.who.int/iris/bitstream/10665/185295/1/WHO_ME RS LAB 15.1 fre.pdf?ua=1
- 9. Alagaili, A. N., T. Briese, N. Mishra, V. Kapoor, S. C. Sameroff, P. D. Burbelo, E. de Wit, V. J. Munster, L. E. Hensley, I. S. Zalmout, A. Kapoor, J. H. Epstein, W. B. Karesh, P. Daszak, O. B. Mohammed, and W. I. Lipkin, 2014: Middle East respiratory syndrome coronavirus infection in dromedary camelsin Saudi Arabia. MBio 5, e00884-00814.
- 10. Chan JFW, Lau SKP, To KKW, Cheng VCC, Woo PCY, Yuen KY (2015) Middle East respiratory syndrome coronavirus: another zoonotic betacoronavirus causing SARS-like disease. Clin Microbiol Rev 28(2): 465–522.https://doi.org/10.1128/CMR.00102-14
- 11. Nowotny N, Kolodziejek J. Middle East respiratory syndrome coronavirus (MERS-CoV) in dromedary camels, Oman, 2013. Euro Surveill. 2014;19(16):pii=20781.
- 12. Hemida, M. G., D. K. Chu, L. L. Poon, R. A. Perera, M. A.Alhammadi, H. Y. Ng, L. Y. Siu, Y. Guan, A. Alnaeem, and M. Peiris, 2014: MERS coronavirus in dromedary camel herd, Saudi Arabia. Emerg. Infect. Dis. 20, 1231–1234.
- 13. Reusken CB, Ababneh M, Raj VS, Meyer B, Eljarah A, Abutarbush S, Godeke GJ, Bestebroer TM, Zutt I, Müller MA, Bosch BJ, Rottier PJ, Osterhaus AD, Drosten C, Haagmans BL, Koopmans MP. Middle East Respiratory Syndrome coronavirus (MERS-CoV) serology in major livestock species in an affected region in Jordan, June to September 2013. Euro Surveill. 2013;18(50):pii=20662. Available online: http://www.eurosurveillance.org/ViewArticle.aspx?ArticleId=20662
- 14. Chantal B.E.M. Reusken, Lilia Messadi, Ashenafi Feyisa, Hussaini Ularamu, Gert-Jan Godeke, Agom Danmarwa, Fufa Dawo, Mohamed Jemli, Simenew Melaku, David Shamaki, Yusuf Woma, Yiltawe Wungak, Endrias Zewdu Gebremedhin, Ilse Zutt, Berend-Jan Bosch, Bart L. Haagmans, and Marion P.G. Koopmans Geographic Distribution of MERS Coronavirus among Dromedary Camels, Africa. Volume 20, Number 8—August 2014
- 15. Reusken, C. B., M. Ababneh, V. S. Raj, B. Meyer, A. Eljarah, S. Abutarbush, G. J. Godeke, T. M. Bestebroer, I. Zutt, M. A.Muller, B. J. Bosch, P. J. Rottier, A. D. Osterhaus, C. Drosten, B. L. Haagmans, and M. P. Koopmans, 2013a: Middle East Respiratory Syndrome coronavirus (MERS CoV) serology in major livestock species in an affected region in Jordan, June to September 2013. Euro. Surveill. 18, 20662.