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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

### *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)**

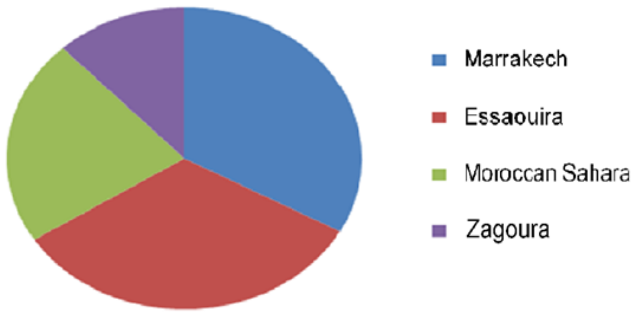
| Sex | Number | Average of age | Extremes |
|---|---|---|---|
| Males | 19 | 3.64 | From 2 weeks to 17 years |
| Females | 81 | 7.2 | From 2 weeks to 16 years |
| **Total** | **100** | **6.59** | From 2 weeks to 17 years |

**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.

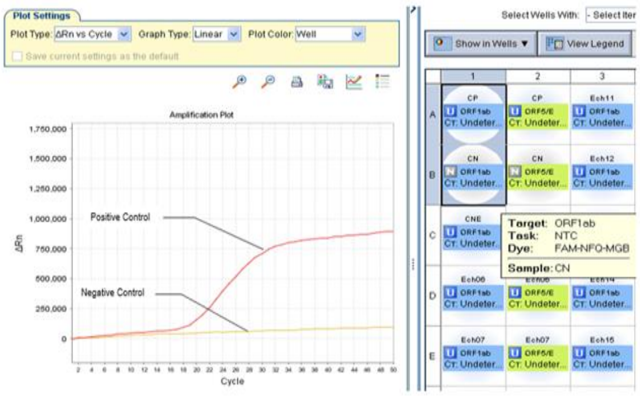
The geographical origin of these dromedaries has shown that 33% of them are from the Essaouira region, 33% from Marrakech, 22% from the Moroccan South and 12% from Zagoura. Dromedaries from these regions may all originate from several regions of the south, as 92% of the camelin herd is located in southern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

**Figure 1:** Geographical distribution of dromedaries.



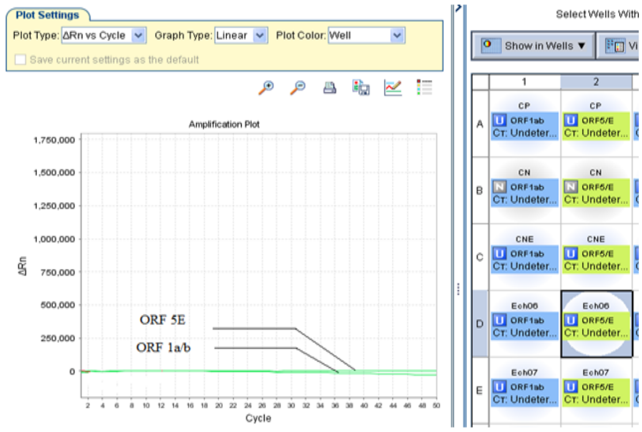
None of the 100 dromedaries analyzed by RT-PCR did not reveal the presence of MERS-Cov, whether for nasal swabs or for stool specimens. **(Figure 2, 3)**

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


Plot Type: ΔRn vs Cycle Graph Type: Linear Plot Color: Well

Target: ORF1ab
Task: NTC
Dye: FAM-NFQ-MGB
Sample: CN

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


Plot Type: ΔRn vs Cycle Graph Type: Linear Plot Color: Well

## 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.

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.

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