

## ORIGINAL BREU

# Respiratory tract infections caused by Human Coronavirus (HCoVs) in Balearic Islands, 2014

*Infecciones del tracto respiratorio causadas por los Coronavirus Humanos (HCoVs) en las Islas Baleares, 2014*

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## Abstract

**Introduction:** The main human Coronavirus (HCoVs) involved in human respiratory tract infections are the 229E, NL63, and OC43. Due to the lack of information about these infections in our country, it seemed important to know its impact on acute respiratory disease.

**Material and Methods:** In the period February-April 2014, we have studied the presence of different respiratory viruses in 950 samples (throat swabs and/or nasopharyngeal aspirates) belonging to 674 (70.9%) children (<15 years) and 276 adults (29.1%). The detection of respiratory viruses was performed using commercial automatic real-time PCR system (Anyplex RV16), detecting 16 different viruses.

**Results:** The overall HCoV detected was 4.6% of all samples studied and 7.8% (44 cases) of the positive samples. The 44 HCoVs detected corresponded to 20 HCoV-OC43 (45.4%), 17 HCoV-NL63 (38.6%) and 7 HCoV-229E (15.9%). The HCoVs alone were detected in 25 cases (56.8%) and with other respiratory viruses in 19 (43.2%) cases (coinfections). In the mixed infections, rhinovirus was detected in 11 cases (57.8%), influenza virus type B in 6 cases (31.5%), adenovirus in 1 case and RSV-A in 1 case. HCoVs were detected in the 26 children (59%) and 18 adults (41%). Of the 44 cases, 13 (29.5%) required hospital admission, No patients infected by HCoVs died as a direct result of respiratory tract infection.

**Conclusions:** This study demonstrates the importance of respiratory infections caused by HCoVs, especially in children

**Keywords:** Human Coronavirus (HCoVs), OC43, NL63, 229E, Epidemiology

## Resumen

**Introducción:** Los principales coronavirus humanos (HCoVs) causantes de infección respiratoria son el 229E, NL63 y OC43. Debido a la falta de información sobre este tipo de infecciones en nuestra comunidad, nos ha parecido importante conocer su impacto en la enfermedad respiratoria aguda.

**Material y método:** En el período Febrero-Abril de 2014 se ha estudiado la presencia de los diferentes virus respiratorios en 950 muestras clínicas (aspirados nasofaríngeos o frotis faríngeos) pertenecientes a 674 niños (<15 años)(70.9%) y 276 adultos (29.1%). La detección viral se ha realizado mediante una RT-PCR comercial que detecta de forma simultánea y diferencial 16 virus distintos (Anyplex RV16).

**Resultados:** El porcentaje global de detección de los HCoVs fue del 4.6% en todas las muestras estudiadas y del 7.8% (44 casos) en las muestras positivas. Se detectaron 20 OC43 (45.4%), 17 NL63 (38.6%) y 7 229E (15.9%). En 25 casos sólo detectó un HCoV (56.8%) y 19 casos (43.2%) la infección fue mixta. En estas infecciones los virus detectados fueron: rinovirus 11 casos (57.8%), virus gripal tipo B en 6 casos (31.5%), y adenovirus y VRS-A un caso cada uno. Los HCoVs se detectaron en 26 niños (59%) y 18 adultos (41%). De los 44 casos, 13 (29.5%) precisaron de ingreso hospitalario. Ningún paciente falleció como consecuencia de la infección respiratoria por el HCoV.

**Conclusiones:** Este estudio demuestra la importancia de las infecciones respiratorias causadas por los coronavirus humanos, especialmente en niños.

**Palabras clave:** Coronavirus humanos (HCoVs), OC43, NL63, 229E, Epidemiología.

## Introduction

Coronaviruses are among the enveloped RNA viruses that are transmitted through the respiratory tract and have been taxonomically classified into three antigenic groups. The main human Coronavirus (HCoVs) involved in respiratory tract infections are the 229E and NL63, belonging to antigenic group 1 (*Alphacoronavirinae*) and OC43 belonging to antigenic group 2 (*Betacoronavirinae*)<sup>1</sup>. The recent detection of a new HCoV (nHCoV-London) respiratory demonstrated the clinical significance of this group of viruses<sup>2</sup>. The incidence of respiratory infections by these viruses varies widely by geographic area. Due to the lack of information about these infections in our country, it seemed important to know its impact on respiratory disease.

The aim of this study is to determine the incidence and epidemiological characteristics of respiratory tract infections caused by HCoV (OC43, 229E and NL63). We investigated the presence of these viruses in respiratory samples in our region (Mallorca, Balearic Islands, Spain).

## Material and methods

### Sample Collection

In the period February-April 2014, we have studied the presence of different respiratory viruses in 950 samples (throat swabs and/or nasopharyngeal aspirates) belonging to 674 (70.9%) children (<15 years) and 276 adults (29.1%).

### RNA and DNA extraction and real-time RT-PCR

The detection of respiratory viruses was performed using commercial automatic extraction system. Nucleic acids were extracted from 700 µl of specimens by easyMAG (Nimbus, Hamilton Robotics, USA). The final elution volume of each sample was 50 µl. The cDNAs were synthesized from extracted RNAs with the cDNA Synthesis Premix (Seegene, South Korea). Respiratory virus detection kit-A and B were used to detect 14 types of RNA viruses and two types of DNA viruses, according to the manufacture's instructions. The amplification and detection process were realized with the CFX96 real-time PCR detection system (Bio-Rad, CA).

## Results

In 557 (58.6%) of the 950 samples studied was detected some kind of respiratory virus. The overall HCoV detected was 4.6% of all samples studied and 7.8% (44 cases) of the positive samples. The 44 HCoVs detected corresponded to 20 HCoV-OC43 (45.4%), 17 HCoV-NL63 (38.6%) and 7 HCoV-229E (15.9%). The HCoVs alone were detected in 25 cases (56.8%) and with other respiratory

viruses in 19 (43.2%) cases (coinfections). In the mixed infections, rhinovirus was detected in 11 cases (57.8%), influenza virus type B in 6 cases (31.5%), adenovirus in 1 case and RSV-A in 1 case. The coinfections of the HCoV-OC43 were detected in 11 cases (55%), the HCoV-NL63 in 8 (47%) cases, and HCoV-229E in any case.

HCoVs were detected in the 26 children (59%) and 18 adults (41%), corresponding to 28 men (63.6%) and 16 women (36.4%). The 20 HCoV-OC43 were detected in 13 children (65%), having an age range of 2 months-7 years and a median age of 24.5 months; and in 7 adults with a range of 22-74 years (median age of 51.8 years). The 17 HCoV-NL63 were detected in 10 children (58.8%) with an age range of 19 days-15 years and a median age of 32.1 months; and in 7 adults (41.2%) with a range of 42-84 years and a median age of 67.4 years. The 7 HCoV-229E were detected in 3 children (42.8%), with a range of 1 month-14 years and a median age of 96.3 months; and in 4 adults (57.2%) with a range of 39-53 years and a median age of 45 years.

Of the 44 cases, 13 (29.5%) required hospital admission, 4 (30.7%) by HCoV-OC43 (20% of all cases), 5 (38.4%) for HCoV-NL63 (29.4% of all cases) 4 and HCoV-229E (57% of all cases). No patients infected by HCoVs died as a direct result of respiratory tract infection.

## Discussion

The HCoVs are responsible for a variable percentage of respiratory tract infections in children and adults. Chinese studies have shown that these viruses can be detected in 4.4% of the children studied, of whom NL63 represented 2.6%, OC43 the 1.5% and 229E the 0.3%<sup>3</sup>. In our study, only in three months, we detected 7.8% of HCoVs, of which 3.5% were OC43, 3.1% NL63 and 1.2% 229E. In the study by Bastien et al.<sup>4</sup>, in Canada, the HCoV-NL63 was detected in 3.6% of the samples studied, this rate is similar to that detected in our study. The study by Gaunt et al.<sup>1</sup>, in The Netherlands, shows a rate of HCoVs circulation between 3.17% and 2.96%, depending on the season of the year. We agree with this study, it seems that these infections occur preferentially in children (59%) and boys (63.6%). Our detection rate is higher, probably due to the winter season analyzed (highest incidence).

One of the problems of interpretation of the meaning of the HCoVs in respiratory disease is the high rate of coinfections. In our study we detected 43.2% of mixed infections, preferably by rhinovirus. Other studies have reported percentages varying between 11-42%, predominantly different respiratory viruses depending on the time of year when the analysis is performed<sup>1,3-5</sup>. We no detected clinical and epidemiological differences between single infections and coinfections with other respiratory viruses.

We are in agreement with Gaunt et al.<sup>1</sup> the infection of HCoVs with another respiratory virus does not affect the ability of these viruses to establish infection. So in mixed infections, detection of HCoVs should not be considered as an accidental infection that does not contribute to the pathogenesis of respiratory infection.

The HCoV-OC43 seems to be the most frequently detected<sup>1,3</sup>. In our study we also show that this virus, with 45.4%, is more frequently detected, especially in children. The 29.5% of our patients required hospital admission due to respiratory disease or underlying disease. It should be noted that 57.1% of all infections caused by HCoV-229E required hospital admission. In these cases all the admissions corresponding to the 4 adult patients, all of whom were immunocompromised, coinciding with the previous study of Gaunt et al.<sup>1</sup>.

This study demonstrates the importance of respiratory infections caused by HCoVs, especially in children. They are one of the leading causes of respiratory urgent consultation and determine a near 30% hospital admission for these pathologies. The new multiplex genome amplification techniques allow a better understanding of the epidemiology of this new type of respiratory infections

## Conflict of interest

We declare that we have no conflict of interest with respect to this study.

## Funding

None declared.

## References

1. Gaunt ER, Hardie A, Claas ECJ, Simmonds P, Templeton KE. Epidemiology and clinical presentations of the four human coronaviruses 229E, HKU1, NL63, and OC43 detected over 3 years using a novel multiplex real-time PCR method. *J Clin Microbiol* 2010; 48:2940-7.
2. Palm D, Pereyaslov D, Vaz J, Broberg E, Zeller H, Gross D et al. Laboratory capability for molecular detection and confirmation of novel coronavirus in Europe, November 2012. *Euro Surveill* 2012; 17(49). doi:pii20335.
3. Chiu SS, Chan KH, Chu KW, Kwan SW, Guan Y, Poon LLM et al. Human coronavirus NL63 and other coronavirus infections in children hospitalized with acute respiratory disease in Hong Kong, China. *Clin Infect Dis* 2005; 40:1721-9.
4. Bastien N, Anderson K, Hart L, Van Caesele P, Brandt K, Milley D et al. Human coronavirus NL63 infection in Canada. *J Infect Dis* 2005; 191:503-6.
5. Wu PS, Chang LY, Berkhout B, van der Hoek L, Lu CY, Kao CL et al. Clinical manifestations of human coronavirus NL63 infection in children in Taiwan. *Eur J Pediatr* 2008; 167:75-80.