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Human bocavirus commonly involved in multiple viral airway infections

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Abstract

Background: Human bocavirus (HBoV) was recently discovered in children with acute respiratory tract infections. We have included a PCR for HBoV in a study on airway infections in children.
Objectives: To study the occurrence of HBoV in Norwegian children, and to evaluate the results of a semiquantitive PCR.
Study design: During a 4-month period in the winter season 2006/2007 we collected nasopharyngeal aspirations from children who were admitted to the Department of Pediatrics. All samples were examined for 17 agents with real-time PCR.
Results: HBoV was detected in 45 of 376 samples (12%). The occurrence of HBoV was stable during the study period. Multiple viral infections were present in 78% of the samples (42% double, 20% triple and 16% quadruple infections). RS-virus, enterovirus and human metapneumovirus were the most frequently codetected agents. In samples with a high load for HBoV, significantly fewer multiple infections were found than in the other samples. Eighty-eight percent of the 25 patients with HBoV recorded as either the only or the dominating virus, and 50% of the other patients, had lower respiratory tract infection. The difference was statistically significant.
Conclusions: HBoV was frequently detected in nasopharyngeal aspirates from children with airway infections in Norway. Multiple viral infections were common among the HBoV-infected patients. Semiquantitive PCR results may be useful for interpretation of clinical relevance.

Keywords: Human bocavirus; Airway; Infection; Multiple; Children

1. Introduction

Human bocavirus (HBoV) was discovered in 2005 (Allander et al., 2005). It belongs to the family *parvoviridae,* and it is the second virus in this family to be associated with human disease (after parvovirus B19). Little is known about the virus' kinetics and cell tropism.

HBoV is common in airway samples from children less than 5 years with respiratory tract infections, and it has a worldwide distribution (Allander et al., 2007; Arnold et al., 2006; Arden et al., 2006; Fry et al., 2007; Manning et al., 2006; Regamey et al., 2007). A causal relationship between HBoV and airway infection has not been established yet, but some controlled studies supporting this hypothesis have been published (Allander et al., 2007; Fry et al., 2007; Kesebir et al., 2006; Maggi et al., 2007; Manning et al., 2006).

We have studied the occurrence of HBoV in Norwegian children with respiratory tract infections, and evaluated the semiquantitative PCR-results against clinical manifestations.

2. Methods

We collected nasopharyngeal aspirates from children who were admitted to the Department of Pediatrics, St. Olavs Hospital, Trondheim University Hospital with respiratory tract infections during the time period November 13, 2006 to March 16, 2007. St. Olavs Hospital is the regional hospital for Mid-Norway covering a population of 640 000.

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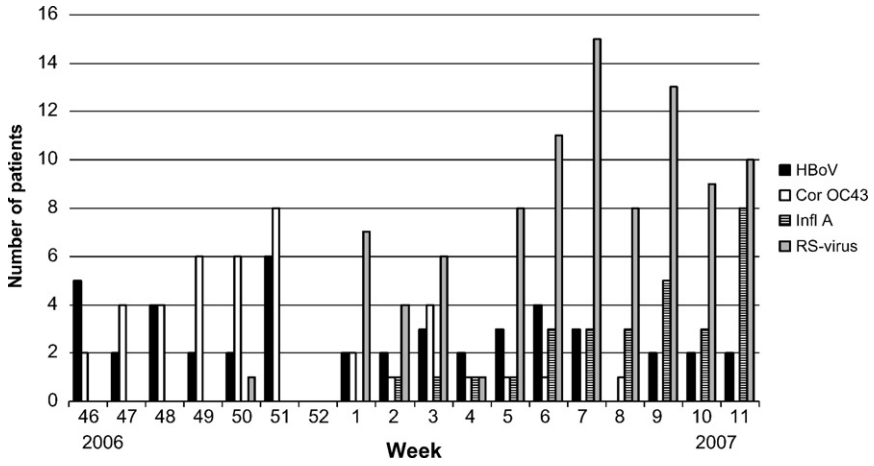

Fig. 1. Number of human bocavirus (HBoV), coronavirus OC43 (Cor OC43), influenza A-virus (Infl A) and RS-virus per week.

Clinical data were obtained from medical records. The children were classified in two main diagnosis categories: lower respiratory tract infection (LRTI) and upper respiratory tract infection (URTI). Other diagnoses not mentioned in the statistics included tonsillitis, gastroenteritis and fever. LRTI was diagnosed in the presence of dyspnea, signs of lower airway obstruction (wheezing, retractions) and/or a positive radiogram (infiltrates, atelectasis, air trapping). URTI was diagnosed when rhinitis, pharyngitis and/or otitis media was present in the absence of signs of LRTI.

Using PCR the nasopharyngeal aspirations were examined for adenovirus, HBoV, coronavirus (OC43, 229E and NL63), enterovirus, human metapneumovirus, influenza A and B virus, parainfluenza virus type 1-3, RS-virus, rhinovirus, *Bordetella pertussis*, *Chlamydia pneumoniae* and *Mycoplasma pneumoniae*. All PCRs were real-time assays based on TaqMan probes. The target for the HBoV-PCR was the NP-1-gene. Selections of primers and probe were based the sequences published by Allander et al. (2005). (Forward primer: CCA CGT GAC GAA GAT GAG CTC, reverse primer TAG GTG GCT GAT TGG GTG TTC, probe CCG AGC CTC TCT CCC CAC TGT GTC G, 5′6-FAM, 3′ TAMRA.) The amount of virus in each sample was recorded semiquantitatively based on the C_t-value (cycle threshold value) and grouped in three categories (high, medium and low viral load). The break points were set to C_t 28 and C_t 35. HBoV was recorded as the dominating virus in a sample when the C_t-value was at least three cycles lower than the C_t-value for any other virus.

In addition all samples were collected on ordinary virus transport media without antibiotics and cultured for viruses and bacteria with standard methods.

3. Results

HBoV was detected in 45 of 376 nasopharyngeal aspirates (12%). It was the fourth most common virus in the material after RS-virus (25%), rhinovirus (17%) and human metapneumovirus (14%). Other common viruses in the material were enterovirus (11%) and coronavirus OC43 (11%). During the 4-month study period the occurrence of HBoV was stable, in contrast to coronavirus OC43, influenza A and RS-virus, which varied significantly (Fig. 1).

At least one virus was detected in 78% of the 376 samples in the total material. One solitary virus was found in 50%,

Table 1
Number of codetected viruses in 35 HBoV-infected patients who had multiple viral infections

	Number of detections			Part of triple/quadruple infections
	Total	PCR	Culture	
Adenovirus	6	5	3	4
Coronavirus OC43	10	10	–	6
Coronavirus NL63	1	1	–	–
Enterovirus	9	9	1	8
Human metapneumovirus	7	7	1	4
Influenza virus A	3	3	3	3
Parainfluenzavirus type 3	5	5	2	3
Rhinovirus	10	10	–	8
RS-virus	8	8	7	3
Cytomegalovirus	2	ND[a]	2	1

The number of detections made by PCR and viral culture are shown. Right column: number of samples with at least two viruses detected in addition to HBoV.
[a] Not done.