World Journal of Pediatrics (2020) 16:293–298
https://doi.org/10.1007/s12519-019-00324-5

ORIGINAL ARTICLE

Human bocavirus in children hospitalized for acute respiratory tract infection in Rome

Laura Petrarca[1] · Raffaella Nenna[1] · Antonella Frassanito[1] · Alessandra Pierangeli[2] · Greta Di Mattia[1] · Carolina Scagnolari[2] · Fabio Midulla[1]

Received: 18 February 2019 / Accepted: 1 November 2019 / Published online: 27 November 2019

Abstract

Background The role of human bocavirus (HBoV) as a respiratory pathogen has not been fulfilled yet. We aimed to describe clinical and serological characteristics of children with HBoV hospitalized for acute respiratory tract infection and to evaluate whether differences occur between HBoV alone and in co-infection.

Methods We retrospectively reviewed data from 60 children (median age of 6.2 months, range 0.6–70.9) hospitalized for acute respiratory symptoms, with HBoV detected from a respiratory sample, using a reverse transcriptase-PCR for 14 respiratory viruses (including respiratory syncytial virus (RSV), influenza virus A and B, human coronavirus OC43, 229E, NL-63 and HUK1, adenovirus, rhinovirus, parainfluenza virus1–3, and human metapneumovirus).

Results HBoV was detected alone in 29 (48.3%) patients, while in co-infection with other viruses in 31 patients (51.7%), with a peak between December and January. Among the 60 patients, 34 were bronchiolitis, 19 wheezing, 3 pneumonia, 2 upper respiratory tract infection, and 2 whooping cough. Seven children (11.6%) required admission to the paediatric intensive care unit (PICU) for respiratory failure. No differences was observed in age, family history for atopy and/or asthma, clinical presentations, chest X-ray, or laboratory findings in children with HBoV alone vs. multiple viral detection. RSV was the most frequently co-detected virus (61.3%). When compared with HBoV detection alone, the co-detection of RSV and HBoV was associated with male sex ($P=0.013$), younger age ($P=0.01$), and lower blood neutrophil count ($P=0.032$).

Conclusions HBoV can be detected alone and in co-infection respiratory samples of children with an acute respiratory tract infection. A cause–effect relationship between HBoV and respiratory infection is not clear, so further studies are needed to clarify this point.

Keywords Human bocavirus · Pediatrics · Respiratory tract infection · Viral infection

Introduction

Human bocavirus (HBoV), a new virus belonging to the *Parvoviridae* family, was first identified in 2005 in respiratory samples of children suffering from viral respiratory infections of unknown etiology [1]. Several studies tried to investigate the role of HBoV, reporting a detection rate ranging from 3.1% [1] to 23.1% [2] in children with acute respiratory tract infections. Sometimes, HBoV was considered responsible for a severe clinical presentation in children with acute respiratory disease [3]; however, other studies reported no differences between HBoV and other viruses in the clinical course of respiratory infections [4].

HBoV is often detected from respiratory samples together with other viruses [5–7], which has been described as a prolonged and intermittent shedding even in asymptomatic patients [3]. In addition, nowadays, the virus does not fulfill the criteria for etiologic association of virus with prevalent disease [8]. As a result, its role of a respiratory pathogen has not been completely accepted yet [9]. To increase our knowledge on the clinical spectrum of HBoV in children with respiratory disease, we aimed to describe the clinical and serological characteristics of children hospitalized for acute respiratory tract infection, whose nasal aspirate or bronchoalveolar lavage (BAL) tested HBoV positive and to investigate whether

✉ Fabio Midulla
midulla@uniroma1.it

[1] Maternal-Infantile and Urological Science Department, "Sapienza" University of Rome, Rome, Italy

[2] Virology Laboratory, Department of Molecular Medicine, "Sapienza" University of Rome, Rome, Italy

there is a difference between single infection and co-infection of HBoV.

Methods

Patients

We retrospectively reviewed clinical records of 60 children (31 males, median age of 6.2 months, range 0.6–70.9) hospitalized for acute respiratory infection in the Pediatric Emergency Department at "Sapienza" University Rome from 2010 to July 2016, in which HBoV was detected from nasal aspirate or BAL. The clinical records included demographic and clinical information [such as age, gender, breastfeeding history, family smoking habit, family history for asthma and atopic diseases, laboratory data including blood neutrophil count, blood lymphocyte count, blood eosinophil count, C- reactive protein (CRP) and the days of hospitalization], chest X-ray records, and the clinical severity score ranging from 0 to 8 that was assigned to each infant on admission in the hospital according to arterial oxygen saturation on room air, presence of retractions, ability to feeding as used in an our previous manuscript [10], and respiratory rate modified according to age as reported for the clinical respiratory score [11].

We classified children into five groups according to clinical, epidemiological, serological, and radiological findings as having:

(1) bronchiolitis: clinically defined as the first episode of acute lower respiratory tract infection in infants up to 12 months of age, characterized by the acute onset of cough, tachypnea, retraction, and diffuse crackles on chest auscultation [12];
(2) preschool wheezing: children aged less than 6 years with lower respiratory tract infection, and wheeze on chest auscultation;
(3) upper respiratory tract infection (URTI), including children with rhinorrhoea and/or cough, without abnormal findings on chest auscultation;
(4) whooping cough, characterized by the presence of cough lasting more than 14 days and at least one of the symptoms of paroxysmal cough, whoop, or post-tussive vomiting with positive laboratory findings for *Bordetella pertussis* [13];
(5) pneumonia, defined as the presence of fever, acute respiratory symptoms, and evidence of a new lung consolidation [14].

HBoV was detected using a reverse transcriptase-PCR (RT-PCR) specific for 14 respiratory viruses including respiratory syncytial virus (RSV), influenza virus (IV) A and B, human coronavirus (HCoV) OC43, 229E, NL-63 and HUK1, adenovirus (AdV), rhinovirus (RV), parainfluenza virus (PiV) 1–3, and human metapneumovirus (MPV) and HBoV, as previously described [15, 16].

The study was approved by the Ethical Committee of Hospital (no.2377/02.02.2012). Researchers adhered to the postulates of the Declaration of Helsinki and an informed consent has been obtained for all patients enrolled.

Statistical analysis

Categorical variables were expressed as numbers and percentages and continuous variables values as median and range or as average and standard deviation. A χ^2 test was performed to compare proportion. The normality of data distribution was tested using the Kolmogorov–Smirnov test. A non-parametric median test (Mann–Whithney *U* test) was performed for the analysis of continuous variables not normally distributed. Two-side P values < 0.05 were considered as statistically significant. Statistical analysis was performed using the SPSS software (version 23.0; SPSS Inc., Chicago, Illinois, USA).

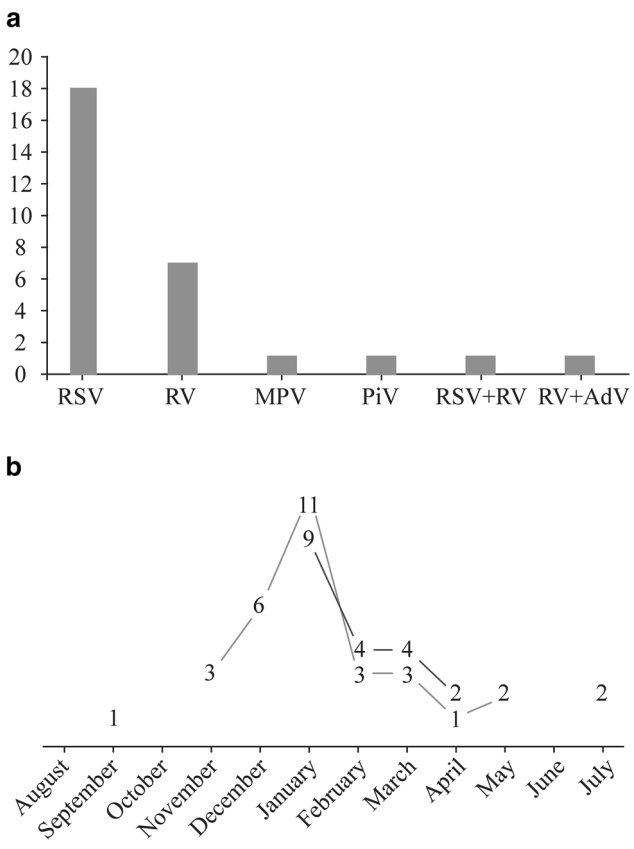

Fig. 1 **a** Absolute numbers of human bocavirus co-detected viruses; **b** monthly distribution of human bocavirus infections. Abbreviation, respiratory syncytial virus (RSV), rhinovirus (RV), human metapneumovirus (MPV), parainfluenza virus (PiV)

Results

Among the 60 children hospitalized for acute respiratory infection, RT-PCR detected HBoV alone in 29 (48.3%) cases and in 31 cases (51.7%) with other viruses. The most frequent co-infections were with RSV and RV (Fig. 1a). Monthly distribution of HBoV showed that 53% of the patients were concentrated in December and January, with no difference between single and co-infection (Fig. 1b).

Among the 60 children in whom HBoV was detected, 34 (56.6%) were hospitalized for bronchiolitis, 19 (31.7%) for wheezing, 3 (5%) for pneumonia, 2 (3.3%) for an upper respiratory tract infection (URTI), and 2 (3.3%) for whooping cough. Clinical, serological, and radiological characteristic, according to diagnosis, are shown in Table 1. HBoV was detected alone in 13 infants with bronchiolitis (38.2%), 11 children with an acute episode of wheezing (57.9%), all the patients with pneumonia and in half with URTI.

In children with whooping cough (diagnosed on the basis of *B. pertussis* detection), HBoV was in co-infection with other viruses in half of the cases (Fig. 2).

Seven children (11.6%) (median age of 11.1 months; range 0.63–44.7) were admitted to the Pediatric Intensive Care Unit (PICU) because of respiratory failure. Among these three children hospitalized for an acute episode of wheezing, HBoV was detected alone; on the contrary, in the four infants with bronchiolitis, HBoV was detected together with RSV. In one child with an acute episode of wheezing, HBoV was detected alone from the BAL. No difference was observed in terms of age, positive family history for atopy and/or asthma, clinical presentation (fever, dyspnea, and food intake reduction) consolidation on chest X-ray, and laboratory findings (CRP, white blood cells count, and lymphocyte count) in children with HBoV detected alone vs. children with multiple viral detection. Children with co-detection were more frequently male ($P=0.019$, by Chi-square test) (Table 2).

Among the children with multiple viral detection, RSV was the most frequently detected virus (61.3%), in which 18 were diagnosed as having bronchiolitis and one upper respiratory tract infection. Infants with HBoV–RSV co-detection appeared more often in male ($P=0.013$), younger ($P=0.01$, by non-parametric median test), and had a lower blood neutrophils count ($P=0.032$) compared with children with HBoV detected alone (Table 3).

When considering only infants with bronchiolitis from RSV to HBoV co-detection, they had a higher clinical severity score (3.5 vs. 2, $P=0.037$) than children with bronchiolitis from HBoV alone and they were more frequently male (15 vs. 6, $P=0.034$). No differences were observed in family history of asthma and/or atopic diseases, breastfeeding, fever, length of hospital stay, CRP, white blood cells count, eosinophils, and lymphocyte count between infants with bronchiolitis from HBoV to HBoV–RSV.

Table 1 Clinical, serological, and radiological characteristic of children hospitalized for respiratory tract infection with human bocavirus divided by diagnosis

Variables	Bronchiolitis ($n=34$)	Wheezing ($n=19$)	URTI ($n=2$)	Pneumonia ($n=3$)	Whooping cough ($n=2$)
Clinical severity score	4.03 ± 2.35	5.58 ± 1.95	1	3	2.5
Clinical severity score ≥ 6	8 (23.5%)	10 (52.6%)	0	0	0
Fever	44.1%	50.0%	0%	100%	0%
Food intake reduction	67.7%	63.1%	100%	0%	100%
Food intake reduction < 75%	12 (35.3%)	10 (52.6)	0	0	0
Total white blood cell count (n/mm^3)	11,986 ± 4400	13,365 ± 5969	24,050	18,790	26,240
Lymphocytes (n/mm^3)	5292 ± 2949	2357 ± 994	3150	4736	9115
Neutrophils (n/mm^3)	4667 ± 3272	9536 ± 5664	20,250	12,078	1960
Eosinophils (n/mm^3)	89 ± 161	232 ± 356	120	70	470
C-RP (mg/dL)	1.7 ± 3.2	1.1 ± 1.1	0.57	0.85	0.03
Chest X-ray					
Consolidation	15/22 (68.2)	10/19 (52.6%)	NA	100%	NA
Air trapping	8/22 (36.4%)	10/19 (52.6)	NA	1 (33.3)	NA
PICU admission	3 (8.8%)	6 (31.6%)	0	0	0
Sodium (mmol/L)	139 (133–145)	139.5 (135–141)	148	139 (137–139)	137
GOT (U/L)	49.12 ± 41.06	35.47 ± 7.06	41	30.33 ± 8.74	31
GPT (U/L)	35.94 ± 15.72	18.33 ± 5.83	16	18.67 ± 9.82	19

C-RP C-reactive protein, *PICU* Paediatric Intensive Care Unit, *GOT* glutamic oxaloacetic transaminase, *GPT* glutamic pyruvate transaminase, *NA* not available

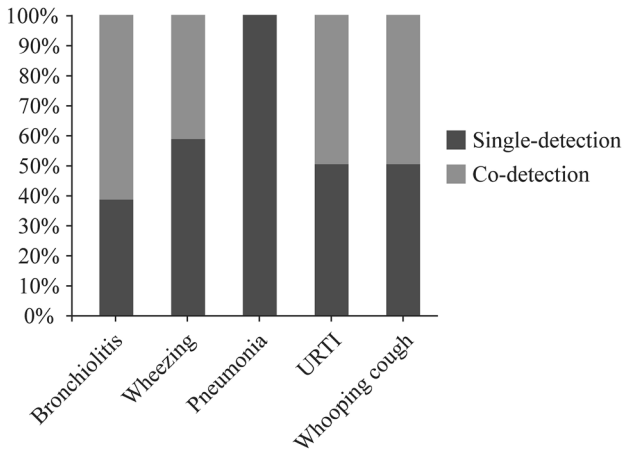

Fig. 2 Frequencies of multiple viruses (dark grey) and single virus (light grey) detection divided according to diagnosis

Discussion

Our study revealed that HBoV could be detected alone or together with other viruses in nasal aspirates and BAL of children hospitalized for acute respiratory tract infection, in 51.7% as a co-detection, with a peak during the winter season.

The frequency of HBoV co-detection is in line with Zhou et al. (52%) and Ljubin-Sternak et al. [2, 17], higher than the one found by Sun et al. [18] and only a bit lower than other studies [5, 7, 19]. In infants hospitalized for an acute respiratory infection, Calvo et al. in one study detected HBoV in combination with other viruses in 64.6% of children and in another study in 75% of the patients [5, 19]. Ghietto et al. confirmed a high rate of HBoV co-detection (78.1%) [7], as well as de Leon et al. (95%) [4] and Martin et al. (72%) [3]. The high rate of co-detection raises doubts on causal relationship of HBoV with respiratory infection, considering its prolonged persistence in the airways likewise. Several authors found the virus in different type of cells [20] and in respiratory samples of healthy volunteers [21]. In addition, we found 3% of HBoV detection among 21 controls without respiratory symptoms in our previous study [16]. The cause–effect relationship seems hard to be approved, but a possible help can rely on HBoV detection from the airways samples during the acute infection together with seroconversion of the anti-HBoV antibody titers [22].

In our series, bronchiolitis was the largest group, and the most frequently HBoV co-detected virus was RSV. In a

Table 2 Clinical, epidemiological, and serological characteristic of children hospitalized for respiratory tract infection with human bocavirus in single vs. co-detection

Variables	Single detection ($n=29$)	Co-detection ($n=31$)	P value
Sex (male)	10 (34.5)	21 (67.7%)	0.019*
Age (mon) median (range)	7.47 (0.7–57.6)	3.63 (0.6–70.9)	0.102
Birthweight (kg) median (range)	3.0 (0.6–4.1)	3.0 (1.8–3.8)	0.684
Breastfeeding (mon) median (range)	1 (0–24)	1 (0–8)	0.839
Breastfeeding	19/28 (67.9)	19/26 (73.1)	0.675
Exposure to smoke	17/27 (63)	14/27 (51.9)	0.409
Maternal smoke during pregnancy	8/27 (29.6)	3/27 (11.1)	0.091
Family history of asthma	9/27 (33.3)	7/28 (25.0)	0.496
Family history of atopy	9/27 (33.3)	9/28 (32.1)	0.925
Fever	14 (50)	13 (41.9)	0.535
Clinical severity score median (range)	4 (0–8)	5 (0–8)	0.308
Days of hospitalization median (range)	4 (2–30)	5 (1–12)	0.367
Admission to PICU	4 (13.8%)	5 (16.1%)	1
Total white blood cell count (n/mm^3) median (range)	12,895 (5010–22,980)	10,830 (5460–36,910)	0.511
Neutrophils (n/mm^3) median (range)	6683 (1001–18,100)	3457 (1079–21,110)	0.168
Lymphocytes (n/mm^3) median (range)	3755 (1310–11,859)	3888 (1090–12,040)	0.511
Eosinophils (n/mm^3) median (range)	75 (0–470)	66 (0–1460)	0.824
Eosinophils > 300/mm^3	4/26 (15.4)	5/26 (17.2)	1
C-RP (mg/dL) median (range)	0.73 (0.03–13.9)	0.8 (0.01–16.98)	0.609
Chest X-ray			
Air trapping	10/22 (45.5)	19/22 (40.9)	0.761
Consolidation	12/22 (54.5)	16/22 (72.7)	0.210

C-RP C-reactive protein, *PICU* Paediatric Intensive Care Unit. *P value by χ^2 test

Table 3 Clinical, epidemiological, and serological findings of children hospitalized for respiratory tract infection with human bocavirus in single vs. human bocavirus and respiratory syncytial virus

Variables	HBoV (n = 29)	RSV + HBoV (n = 18)	*P* value
Sex (male)	10 (34.5)	13 (72.7%)	0.013*
Age (mon) median (range)	7.5 (0.7–57.6)	2.9 (0.6–11.1)	0.001†
Birthweight (kg) median (range)	3.0 (0.6–4.1)	3.0 (1.8–3.8)	0.514
Breastfeeding (mon) median (range)	1 (0–24)	1 (0–5)	0.669
Breastfeeding	19/28 (67.9)	12/16 (75.0)	0.617
Exposure to smoke	17/27 (63)	10/18 (51.9)	0.619
Maternal smoke during pregnancy	8/27 (29.6)	2/18 (11.1)	0.143
Family history of asthma	9/27 (33.3)	5/18 (27.8)	0.693
Family history of atopy	9/27 (33.3)	5/18 (27.8)	0.693
Fever	14 (50)	7 (38.9)	0.460
Clinical severity score median (range)	5 (1–8)	4 (0–8)	0.331
Admission to PICU	4 (13.8%)	3 (16.7%)	1
Days of hospitalization median (range)	4 (2–30)	5 (1–12)	0.128
Total white blood cell count (n/mm^3) median (range)	12,895 (5010–22,980)	10,805 (5460–22,600)	0.086
Neutrophils (n/mm^3) median (range)	6683 (1001–18,100)	2961 (1079–11,179)	0.032†
Lymphocytes (n/mm^3) median (range)	3755 (1310–11,859)	4388 (1455–11,707)	0.145
Eosinophils (n/mm^3) median (range)	75 (0–470)	25 (0–500)	0.438
Eosinophils > 300/mm^3	4/26 (15.4)	2 (11.1)	1
C-RP (mg/dL) median (range)	0.73 (0.03–13.9)	0.7 (0.01–17.0)	0.613
Chest X-ray			
Air trapping	10/22 (45.5)	5/14 (35.7)	0.563
Consolidation	12/22 (54.5)	11/14 (78.6)	0.143

C-RP C-reactive protein, *PICU* pediatric intensive care unit. **P* value by χ^2 test; †*P* value by Mann–Whitney *U* test and non-parametric median test

previous study from our group [9], we found that in infants with bronchiolitis, the RSV–HBoV co-detection was associated with higher severity and longer hospital stay than infants with bronchiolitis caused by RSV or RV or HBoV alone. Our study found that the co-detection of HBoV with RSV was associated with younger age, male sex, and lower neutrophils peripheral count, compared to HBoV detection alone. Considering only infants hospitalized for bronchiolitis, we confirmed our previous findings that the RSV–HBoV co-detection was associated with higher severity of the disease. Other studies confirmed the higher clinical severity presentation of RSV–HBoV co-infection in infants with respiratory infection [18].

A HBoV co-detection was registered in 42.1% patients with an acute episode of wheezing. Viral trigger in preschool wheeze is nowadays universally recognized, both for episodic and multiple trigger of wheezing [23]. Association of HBoV in respiratory samples and wheezing has been largely described in children [4–6, 17]. Calvo et al. found [19] infections caused by HBoV were clinically similar to those by RV, except for a higher necessity of oxygen support in the cases of HBoV. In our series, two out of the 19 infants (10.5%) hospitalized for wheezing had a severe clinical presentation and required admission to the PICU. HBoV was detected alone in severe cases and in all cases of pneumonia, suggesting that HBoV might play the role of a pathogen in patient with severe lower respiratory tract infection.

HBoV had a peak during winter season (December–January), which was similar to the peak of RSV in our country, with no difference between single and co-infection. This trend reflected the distribution of bronchiolitis in our country [24], and it was in line with the previous studies [1, 25]. However, the peak months were in contrast with the data reported by Silva et al., in which HBoV infections occurred more frequently during spring and autumn seasons [26]. Probably, the controversies about the seasonal distribution for HBoV arised from the different population enrolled in the different studies and also from the different geographic areas.

Our retrospective study had several limitations. First, the study lacked healthy controls; second, viral load was not performed on nasal samples, and finally, serological antibodies of HBoV were not assessed.

In conclusion, our study demonstrates that HBoV can be detected in children with acute respiratory tract infection both as single agent and as a co-infection, without any differences regarding the clinical presentation. However,

the cause–effect relationship between HBoV detection and respiratory infections cannot be achieved, yet based on our data and further studies are needed to clarify if HBoV can play a pathogenetic role in respiratory diseases of children.

Author's contribution LP and RN: conception and design of the study; FM, CS, and AP: critical revision of the article for important intellectual content; AF and GDM: patients' enrollment. FM, CS, and AP: critical revision of the article for important intellectual content; AF and GDM: patients' enrollment.

Funding None.

Compliance with ethical standards

Ethical approval This study was approved by Policlinico Umberto I Ethic Committee (no. 2377/02.02.2012).

Conflict of interest The authors declare that they have no conflict of interest.

References

1. Allander T, Tammi MT, Eriksson M, Bjerkner A, Tiveljung-Lindell A, Andersson B. Cloning of a human parvovirus by molecular screening of respiratory tract samples. Proc Natl Acad Sci USA. 2005;102:12891–6.
2. Ljubin-Sternak S, Meštrović T, Ivković-Jureković I, Tešović G, Mlinarić-Galinović G, Lukšić I, et al. High detection rates of human bocavirus in infants and small children with lower respiratory tract infection from Croatia. Clin Lab. 2019. https://doi.org/10.7754/Clin.Lab.2018.180702.
3. Martin ET, Fairchok MP, Kuypers J, Magaret A, Zerr DM, Wald A, et al. Frequent and prolonged shedding of bocavirus in young children attending daycare. J Infect Dis. 2010;201:1625–32.
4. Do Amaral de Leon C, Amantea SL, Pilger DA, Cantarelli V. Clinical and epidemiologic profile of lower respiratory tract infections associated with human bocavirus. Pediatr Pulmonol. 2013;48:1112–8.
5. García-García ML, Calvo C, Falcón A, Pozo F, Pérez-Breña P, De Cea JM, Casas I. Role of emerging respiratory viruses in children with severe acute wheezing. Pediatr Pulmonol. 2010;45:585–91.
6. Calvo C, García-García ML, Pozo F, Carballo D, Martínez-Monteserín E, Casas I. Infections and coinfections by respiratory human bocavirus during eight seasons in hospitalized children. J Med Virol. 2016;88:2052–8.
7. Ghietto LM, Majul D, Ferreyra Soaje P, Baumeister E, Avaro M, Insfrán C, et al. Comorbidity and high viral load linked to clinical presentation of respiratory human bocavirus infection. Arch Virol. 2015;160:117–27.
8. Huebner RJ. Criteria for etiologic association of prevalent viruses with prevalent diseases; the virologist's dilemma. Ann NY Acad Sci. 1957;67:430–8.
9. Williams JV. Déjà vu all over again: koch's postulates and virology in the 21st century. J Infect Dis. 2010;201:1611–4.
10. Midulla F, Scagnolari C, Bonci E, Pierangeli A, Antonelli G, De Angelis D, et al. Respiratory syncytial virus, human bocavirus and rhinovirus bronchiolitis in infants. Arch Dis Child. 2010;95:35–41.
11. Liu LL, Gallaher MM, Davis RL, Rutter CM, Lewis TC, Marcuse EK. Use of a respiratory clinical score among different providers. Pediatr Pulmonol. 2004;37:243–8.
12. Smyth RL, Openshaw PJ. Bronchiolitis. Lancet. 2006;368:312–22.
13. Cherry JD, Grimprel E, Guiso N, Heininger U, Mertsola J. Defining pertussis epidemiology: clinical, microbiologic and serologic perspectives. Pediatr Infect Dis J. 2005;24:S25–34.
14. Harris M, Clark J, Coote N, Fletcher P, Harnden A, McKean M, et al. British thoracic society guidelines for the management of community acquired pneumonia in children: update 2011. Thorax. 2011;66:1–23.
15. Pierangeli A, Gentile M, Di Marco P, Pagnotti P, Scagnolari C, Trombetti S, et al. Detection and typing by molecular techniques of respiratory viruses in children hospitalized for acute respiratory infection in Rome, Italy. J Med Virol. 2007;79:463–8.
16. Pierangeli A, Scagnolari C, Trombetti S, Grossi R, Battaglia M, Moretti C, et al. Human bocavirus infection in hospitalised children in Italy. Influenza Other Respir Viruses. 2008;2:175–9.
17. Zhou L, Zheng S, Xiao Q, Zhou L, Zheng S, Xiao Q, et al. Single detection of human bocavirus 1 with high viral load in severe respiratory tract infections in previously healthy children. BMC Infect Dis. 2014;14:424.
18. Sun H, Sun J, Ji W, Hao C, Yan Y, Chen Z, et al. Impact of RSV coinfection on human bocavirus in children with acute respiratory infections. J Trop Pediatr. 2019;65:342–51.
19. Calvo C, Garcìa-Garcìa ML, Pozo F, Carvajal O, Pérez-Breña P, Casas I. Clinical characteristic of human bocavirus infections compared with other respiratory viruses in Spanish children. Pediatr Infect Dis J. 2008;27:677–80.
20. Lüsebrink J, Schildgen V, Tillmann RL, Wittleben F, Böhmer A, Müller A, et al. Detection of head-to-tail DNA sequences of human bocavirus in clinical samples. PLoS ONE. 2011;6:e19457.
21. Byington CL, Ampofo K, Stockmann C, Adler FR, Herbener A, Miller T, et al. Community surveillance of respiratory viruses among families in the utah better identification of germs-longitudinal viral epidemiology (BIG-LoVE) study. Clin Infect Dis. 2015;61:1217–24.
22. Kantola K, Hedman L, Allander T, Jartti T, Lehtinen P, Ruuskanen O, et al. Serodiagnosis of human bocavirus infection. Clin Infect Dis. 2008;46:540–6.
23. Brand PLP, Caudri D, Eber E, Gaillard EA, Garcia-Marcos L, Hedlin G, et al. Classification and pharmacological treatment of preschool wheezing: changes since 2008. Eur Respir J. 2014;43:1172–7.
24. Cangiano G, Nenna R, Frassanito A, Evangelisti M, Nicolai A, Scagnolari C, et al. Bronchiolitis: analysis of 10 consecutive epidemic seasons. Pediatr Pulmonol. 2016;51:1330–5.
25. Pozo F, García-García ML, Calvo C, Cuesta I, Pérez-Breña P, Casas I. High incidence of human bocavirus infection in children in Spain. J Clin Virol. 2007;40:224–8.
26. Silva PE, Figueiredo CA, Luchs A, de Paiva TM, Pinho MAB, Paulino RS, et al. Human bocavirus in hospitalized children under 5 years with acute respiratory infection, São Paulo, Brazil, 2010. Arch Virol. 2018;163:1325–30.