MINI-REVIEW

The prevalence of human bocavirus in <2-year-old children with acute bronchiolitis

S. Falahi1, H. Sayyadi2, A. Abdoli3, A. Kenarkoohi4 and S. Mohammadi5

1) Zoonotic Diseases Research Center, 2) Department of Biostatistics, Faculty of Health, Ilam University of Medical Sciences, Ilam, 3) Department of Parasitology and Mycology, School of Medicine, 4) Zoonoses Research Centre, Jahrom University of Medical Sciences, Jahrom, 5) Department of Microbiology, Faculty of Medicine and 6) Department of Operating Room, School of Allied Medical Sciences, Ilam University of Medical Sciences, Ilam, Iran

Abstract

Acute bronchiolitis is one of the most common lower respiratory tract infections in children with less than 2 years of age. Nowadays, molecular methods provide an opportunity to better understand the etiology of bronchiolitis. Several viral agents including Respiratory syncytial virus (RSV), Rhinovirus, Parainfluenza and Human bocavirus (HBoV) are responsible for acute bronchiolitis. There are growing studies on the prevalence of HBoV in patients with bronchiolitis. The present systematic review and meta-analysis were conducted to determine the pooled prevalence of HBoV in the respiratory samples of children with acute bronchiolitis. A literature search was conducted in the databases of PubMed, Scopus and Web of Science to recruit studies reporting the frequency of HBoV in <2-year-old children with acute bronchiolitis from 2005 to 2019. Only studies that used polymerase chain reaction (PCR)-based methods to detect the virus in nasopharyngeal samples were included. A total of 22 studies assessing 6751 cases were analyzed. According to the meta-analysis based on the random-effects model, the overall prevalence of HBoV in children with <2 years old was obtained 13% (95% CI: 0.09-0.17). Additionally, the rates of single (as the sole organism) and mixed (in combination with other viruses) HBoV infections were 4% and 9%, respectively. This study showed a high rate of HBoV detection in children with acute bronchiolitis. This should be considered as part of a diagnostic test panel for respiratory infections in children with bronchiolitis.

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Introduction

Bronchiolitis is a common respiratory tract infection in children. It is associated with inflammation, edema, increased mucus production, and the necrosis of the epithelial cells of bronchioles [1]. Bronchiolitis primarily presents with the symptoms of upper respiratory tract infections. These include nasal congestion, fever, and cough progressing to wheezing and tachypnea. Because the lungs and immune system of young children are not fully maturated, acute bronchiolitis can be a serious clinical condition leading to the blockage of airways in these children. Acute bronchiolitis is generally diagnosed based on clinical and chest X-ray (CXR) examinations; however, complete blood cell count (CBC) can also be helpful [2].

Several respiratory viruses are known to cause bronchiolitis, among these, respiratory syncytial virus (RSV) is responsible for 70% of the cases. Other causative agents include human rhinovirus (HRV), type A and B influenza viruses, parainfluenza, adenovirus, human coronavirus, and human bocavirus (HBoV) which is a relatively new etiology of bronchiolitis.

Studies on the pathogenic role of HBoV in humans are on the rise [3]. HBoV is a non-enveloped DNA virus that belongs to the Parvoviridae family, the subfamily of Parvovirinae, and the genus of Bocavirus. This virus was first discovered by Allander et al., and phylogenetically, it reveals similarities with...
parvoviruses from bovine and canine [4]. HBoV, which has been detected in respiratory, fecal, urine, saliva, and blood specimens, shows a global distribution [5,6]. After the identification of HBoV, many studies have reported this virus as a cause of acute respiratory diseases in children. However, the frequent co-detection of HBoV with other viruses in patients’ samples has debated its role as a true pathogen, and some suggested the virus as a passenger agent [7,8]. On the other hand, although HBoV persistence is not fully understood, HboV DNA remains in the tissues of children’s respiratory tract for a long time after the primary infection, it may be one reason for identifying HBoV as a co-infection in some studies [7,9,10].

So far, there has been one fatality reported in an immunocompetent child due to severe acute bronchiolitis caused by HBoV [11]. Some studies have described the HBoV as the second or third most prevalent cause of bronchiolitis [3,12]. The seasonal distribution of HBoV is similar to that of RSV, leading to a prominent coincidence between these two infections [13]. Overall, the prevalence of HBoV has been widely variable in patients with bronchiolitis in different studies [14].

Here, we conducted a systematic review and meta-analysis to estimate the frequency of HBoV in < 2-year-old children with acute bronchiolitis.

Materials and methods

Searching and selection of related articles
According to the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) recommendations [15], we searched various databases including PubMed, Scopus and Web of Science for related articles using the Medical Subject Heading (MeSH) terms of “bocavirus”, “HBoV”, “prevalence”,

TABLE 1. The characteristics of the studies included in this review

| Author          | Year | Country | Numer of case | Male | Female | Sample type          | Mean age (months) | Number of positive | Percent | Coinfection | Single infection |
|-----------------|------|---------|---------------|------|--------|----------------------|-------------------|-------------------|---------|-------------|-----------------|
| Zappa A et al.  | 2011 | Italy   | 22            | NA   | NA     | Pharyngeal Swabs     | 5                 | 3                 | 12.4    | NA          | NA              |
| Uyar M et al.   | 2014 | Turkey  | 62            | 45   | 17     | Nasopharyngeal Aspirate | N             | 3                 | 4.8     | 2           | 1               |
| Ricart S et al. | 2013 | Spain   | 404           | NA   | NA     | Nasopharyngeal Aspirate | 3.6              | 118               | 18.8    | 101          | 17              |
| Brand HK et al. | 2012 | Netherlands | 142          | NA   | NA     | Nasopharyngeal Aspirate | 4.5              | 6                 | 4.22    | 6           | 0               |
| Calvo C et al.  | 2010 | Spain   | 318           | 175  | 193    | Nasopharyngeal Aspirate | 5.6              | 42                | 11.4    | 28          | 14              |
| Chen yw et al.  | 2014 | Taiwan  | 113           | 77   | 36     | Nasopharyngeal Aspirate | 9                 | 22                | 19.5    | 16          | 6               |
| Chung Y et al.  | 2008 | Republic of Korea | 308         | NA   | NA     | Nasopharyngeal Aspirate | 11.4             | 37                | 12      | 8           | 29              |
| Rosal TD et al. | 2015 | Spain   | 684           | NA   | NA     | Nasopharyngeal Aspirate | 9.1              | 31                | 4.5     | 0           | 31              |
| Pierangeli A et al. | 2008 | Italy   | 204           | 102  | 102    | Nasal Washings       | N              | 22                | 10.7    | 16          | 6               |
| Jacques J et al. | 2008 | France  | 192           | NA   | NA     | Nasopharyngeal Aspirate | 8.6              | 24                | 12.5    | 10          | 14              |
| Midulla P et al. | 2010 | Italy   | 182           | NA   | NA     | Nasal Washings       | N              | 22                | 12.5    | 15          | 7               |
| Ricart S et al. | 2013 | Spain   | 484           | 235  | 173    | Nasopharyngeal Aspirate | 12               | 119               | 24.58   | NA          | NA              |
| Wang k et al.   | 2009 | China   | 51            | NA   | NA     | Nasopharyngeal Aspirate | N              | 11                | 21.6    | NA          | NA              |
| Chen ZR et al.  | 2014 | China   | 998           | 672  | 326    | Nasopharyngeal Aspirate | 8.5              | 82                | 11.6    | 24          | 58              |
| Robledo MA et al. | 2018 | Mexico  | 134           | 81   | 53     | Nasopharyngeal Aspirate | 8.6              | 9                 | 5.9     | 0           | 9               |
| Janahi I et al. | 2017 | Qatar   | 369           | 247  | 122    | Nasopharyngeal Aspirate | N              | 15                | 4.1     | NA          | NA              |
| Cangiano G et al. | 2016 | Italy   | 723           | 395  | 201    | Nasopharyngeal Aspirate | 2.13             | 13                | 1.8     | 13          | 0               |
| Przanik A et al. | 2018 | Slovenia | 473          | NA   | NA     | Nasopharyngeal Swabs  | 10               | 87                | 18.4    | NA          | NA              |
| Mascio P et al. | 2011 | Portugal | 78            | 47   | 30     | Nasopharyngeal Aspirate | 8.5              | 29                | 37.1    | 26          | 3               |
| Huguenin A et al. | 2012 | France  | 138           | 81   | 57     | Nasopharyngeal Aspirate | 4                 | 37                | 27      | 24          | 13              |
| Antoni F et al. | 2010 | Portugal | 207          | NA   | NA     | Nasopharyngeal Aspirate | N              | 8                  | 3.8     | 6           | 2               |
| Miron D et al.  | 2010 | Israel  | 465           | 279  | 186    | Sputum or Nasal Wash | 3                 | 31                | 6.7     | 28          | 3               |

NA: Not Available.
“frequency”, “epidemiology”, “acute bronchiolitis”, “lower respiratory tract infection”, and “acute respiratory tract infection” either alone or in combination with each other applying the Boolean operators (AND, OR).

All the related papers published from January 2005 to October 2019 were retrieved. After removing duplicates, the abstracts and full texts of the studies were reviewed. In addition, to make sure that the search was complete, the reference lists of eligible articles were also manually searched for related works. Furthermore, the links to “similar articles” on the PubMed had been opening to find other relevant papers.

Inclusion and exclusion criteria
All English-language research articles published on the prevalence of HBoV in children with acute bronchiolitis were enrolled. Other types of articles including reviews and letters to editors were excluded. In addition, studies that used non-molecular methods for detecting the virus were excluded. Studies performed on children older than 2 years were removed from the assessment.

Data extraction
The following data was collected and recorded into a checklist: first authors’ names, year of publication, the country of study conduction, the number of cases, the number of male and female subjects, sample types, patients’ mean age, the number and percentage of positive samples, and finally the rates of single (only HBoV) and mixed (HBoV along with other pathogen) infections.

Statistical analysis
To analyze and combine the results of different studies, a binomial distribution was considered for the prevalence of the virus in each study. The standard errors were calculated based on this distribution. Heterogeneity was assessed using the Cochran Q test and I² index. Based on the high heterogeneity rate among the studies, the random effects model was used for meta-analysis. Meta-regression analysis was applied to investigate the sources of heterogeneity and the relationship between the virus prevalence and each of sample size and year of publication. Sensitivity analysis was also used to investigate the impact of each study on the calculation of the final outcome. All the analyses were performed in STATA 11 software, and P values less than 0.05 were considered significant in all the statistical tests.

Results
A total of 2936 articles were collected upon the initial search in the electronic databases. Of these, 722 duplicate studies were removed. By subsequent screenings, 1814 additional articles...
were excluded according to our exclusion criteria. After that, the full texts of 400 articles were carefully assessed. Of these, 378 studies met at least one exclusion criterion. Finally, 22 articles were included in the meta-analysis process (Fig. 1).

In this study, a total of 22 articles comprising of 6751 children with a mean age of 6.7 months (95% CI: 5.1-8.3) were included in the meta-analysis process. The details of the reviewed articles [12,13,16–35] have been shown in Table 1. The rate of heterogeneity among the studies was high (97.4%), and therefore the random effects model was used for meta-analysis. Sensitivity analysis was also used to investigate the impact of each study on the calculation of the final outcome (Fig. 2).

Based on studies conducted from 2005 to 2019, the overall prevalence of HBoV in the airway samples of <2-year-old children with bronchiolitis was estimated as 13% (95% CI: 0.09-0.17) (Fig. 3).

The lowest and highest frequencies of HBoV were reported by Cangiano et al. (1.8%) and Macao et al. (37.1%), respectively (Fig. 4) [27,30].

A meta-regression model was used to investigate the relationship between the prevalence of HBoV in children and each of the year of study and sample size. Fig. 4 shows the relationships between the prevalence of HBoV and the year of study conduction (Fig. 4A) and sample size (Fig. 4B). Although there were decreasing trends in the prevalence of HBoV by elevations in the year and sample size, with respect to the obtained p values and regression coefficients (0.444 and 0.235, respectively), these associations were not statistically significant.

**FIG. 3.** The prevalence of HBoV in nasopharyngeal aspirate specimens of < 2-year-old children with bronchiolitis based on the random effects model. The points represent percentages, and the lengths of lines show 95% confidence interval in each study. The rhombic symbol indicates the overall prevalence of HBoV in all the studies.

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Prevalence of HBoV as either a mono- or mixed-infection

For calculating the prevalence of single HBoV infection in <2-year-old children with bronchiolitis, a total of 15 articles were included in the meta-analysis process. The total number of children in these studies was 4487 with an average age of 6.7 months (95% CI: 5.1-8.3). As the rate of heterogeneity among the studies was high (83.2%), the random effects model was used for meta-analysis.

As shown in Fig. 5, the prevalence of HBoV as a single infection in <2-year-old children with bronchiolitis was 4.4% (95% CI: 3.0-5.7, Fig. 5). The prevalence of HBoV as a co-infection in these children, which was estimated based on studies on 6751 children with an average age of 6.7 months, was 8% (95% CI: 0.04-0.12, Fig. 6).

Discussion

Bronchiolitis is a major cause of hospitalization due to respiratory infections among children with the age of <6 months. Several viral agents including respiratory syncytial virus, HRV, influenza, and human metapneumovirus have been associated with bronchiolitis in children. In addition to the above-mentioned viruses, HBoV has also been reported as a cause of bronchiolitis; however, this association is still controversial. In fact, some studies have described a considerably wide range of HBoV infection in patients with bronchiolitis [36,37].

The findings of our study estimated an overall prevalence of 13% for HBoV infection in <2-year-old children with bronchiolitis. These results support previous studies in which HBoV...
has been frequently detected in the samples of patients with bronchiolitis, sometimes even as the second or third most common viral agent [12].

The rate of HBoV infection in children with bronchiolitis varies from 1.8% to 37% in different countries [27,30]. Among the studies assessed here, the lowest and highest frequencies of HBoV were reported by Cangiano et al. (1.8%) in Italy and Macao et al. in Portugal (37.1%). This indicates that geographical location may be one of the factors contributing to the heterogeneity observed among the analyzed studies. In accordance, the prevalence of HBoV has been variable between different countries, and even within countries [27,30].

Most of the assessed studies had been conducted in Spain and Italy. Four studies in Italy from 2005 to 2016 reported frequencies from 1.8% to 12.5% [13,27]. Our findings also suggested that the prevalence of HBoV may be associated with the year of study conduction. Another study conducted in 2016 reported a HBoV prevalence of 1.8% in children with a mean age of 2.1 months [27]. According to previous studies, the prevalence of HBoV infection is generally higher within the ages of 3 to 6 months [2].

Based on the meta regression model, HBoV prevalence showed a decreasing trend over time. Although this observation was not statistically significant, this declining trend may be related to improved health conditions, educations, and also increasing awareness about the transmission ways of respiratory infections.

Although the overall prevalence of HBoV infection was estimated as 12.9%, the incidence of the virus as a single agent was 4%. This supports previous reports noting that HBoV is more commonly identified as a mixed infection in association with other viruses [8].

Although HBoV infection is diagnosed throughout the year, but it peaks during winter and spring [38]. Nevertheless, the seasonal occurrence of HBoV is still a subject of debate, and there are increasing evidences suggesting the higher frequencies of the viral infection in the cold months of the year, especially in January and February [36]. So, sampling time can also be a factor contributing to the heterogeneity among the studies. In fact, in studies with a longer sampling time (e.g. 1-year period), the prevalence of respiratory infections may be underestimated compared to studies performed during the months showing a peak activity of respiratory viruses. This can actually be a source of bias in these studies. Therefore, this factor should be considered in meta-analysis studies to investigate the prevalence of respiratory viruses.

In this study, although inclusion criteria were applied to select studies on a similar population (i.e. children younger than
2 years) and with same detection methods (i.e. molecular assays), there was still a high heterogeneity among the studies. It seems that several factors such as seasonal distribution, age spectrum, year of study, geographical location, and sampling time may be responsible for the heterogeneity observed among the studies.

From the limitations of this study was a high heterogeneity among the studies. Furthermore, there were no reports in many countries limiting our results to certain geographical locations.

**Conclusion**

The data obtained here shows that HBoV infection is relatively frequent in children with bronchiolitis aged <2 years old. HBoV may be either a cause or a risk factor for bronchiolitis in these children. Therefore, this virus should be considered when determining the etiology of bronchiolitis in young children. It is also recommended to incorporate HBoV testing in bronchiolitis diagnostic panels.

**Conflict of interest**

The authors declare no conflict of interest.

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