First Detected Human Bocavirus in a Malaysian Child with Pneumonia and Pre-existing Asthma: A Case Report

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SUMMARY

Human bocavirus (HBoV) is a newly discovered parvovirus associated with respiratory disease in children. There are many reports worldwide on the endemicity of this virus. Since it is relatively new, detection in clinical laboratories is not routinely performed. We describe the first detection of HBoV in Malaysia in a 13-month-old boy with pneumonia and underlying asthma. The infective agent was confirmed by molecular methods.

KEY WORDS:
Respiratory, Bocavirus, Paediatric, Lower Respiratory Tract Infection, Pneumonia, Asthma, Malaysia

INTRODUCTION

Acute respiratory tract infection (ARTI) is the most common cause of morbidity and mortality among children and a frequent reason of both outpatient visits and hospitalisations. RNA viruses are the most causative agents of the common cold among humans. Human rhinoviruses (HRV), respiratory syncytial virus (RSV) and parainfluenza viruses (PIV) are among the most commonly detected viruses. The lesser known viruses such as human metapneumovirus (HMPV) and members of the genus Coronavirus contribute to respiratory disease which can present as severe illness. In 2005, Allander *et al* [1] reported the discovery of a new virus named HBoV in respiratory secretions from children with respiratory tract infections in Sweden. It has been classified in the family Parvoviridae subfamily Parvovirinae, genus Bocavirus and shares the same family with another well known pathogen, parvovirus B19. Human bocavirus is a non-enveloped, round (19 – 22 nm) and icosahedral in symmetry. The genome is not segmented and the complete genome is 4000-6000 nucleotides long. Since its identification, the viral genome has been detected worldwide in nasopharyngeal swabs, serum and fecal samples. It has been found in young children with respiratory or gastrointestinal symptoms[2].

CASE REPORT

The patient was a 13-month-old Malay boy admitted to Serdang hospital. He presented with a 5- day history of runny nose and fever and a 2- day history of cough prior to admission. He developed rapid breathing with wheezing a day prior to admission. On physical examination, the child was afebrile but tachypneoic (respiratory rate of 42 breaths/min) with intercostal and subcostal recessions. Examination of his throat showed bilateral injected tonsils. Auscultation of the lungs revealed coarse crepitations which were confined to the right upper and lower zones with prolonged expiratory phase. The findings of the other systems were unremarkable.

He was born prematurely at 33 weeks of gestation and was ventilated for the first 24 hours of life for respiratory distress syndrome. There was no history of oxygen dependency during neonatal period. He was discharged well at thirty-five days of life. He was subsequently hospitalised for acute bronchiolitis at the age of two and a half months, presented with one week history of cough and runny nose, fever and rapid breathing prior to admission. At the age of five months, his symptoms recurred and during this admission, further evaluation of the presenting complaint pointed to a diagnosis of infantile asthma supported by previous history of bronchiolitis, a strong family history of asthma and atopy on the paternal side. He was prophylactically started on metered dose fluticortisone 250 micrograms twice daily and salbutamol inhaler, the latter to be used when needed.

The hematological test done on admission showed hemoglobin of 13.0 g/l, leucocytosis of 14 x 10^9/l with no predominance in differential counts. The platelet count was 349,000/µl. The erythrocyte sedimentation rate and haematocrit were normal. The chest radiograph showed infiltration in the right lower zone and treated with erythromycin. He improved and was discharged after two days.

Nasopharyngeal aspirate (NPA) was obtained and tested for the presence of the viral agents. Direct immunofluorescence assay (DFA) (Diagnostic Hybrids, USA) was negative for influenza virus A and B, PIV types 1, 2, and 3, HMPV, adenovirus and RSV. The molecular methods utilized both multiplex and regular PCR to detect influenza types A and B, PIV types 1, 2, 3 and 4, HMPV, RSV A and B, adenovirus, coronaviruses OC43, 229E, HKU11 and NL63, rhinovirus and human bocavirus. The results of all the NPA tests were negative, except for the molecular diagnostic test for human bocavirus (Figure 1). The first primer pair for the detection of HBoV was 5′-GACCTCTGTAAGTACTATTAC-3′ and 5′-

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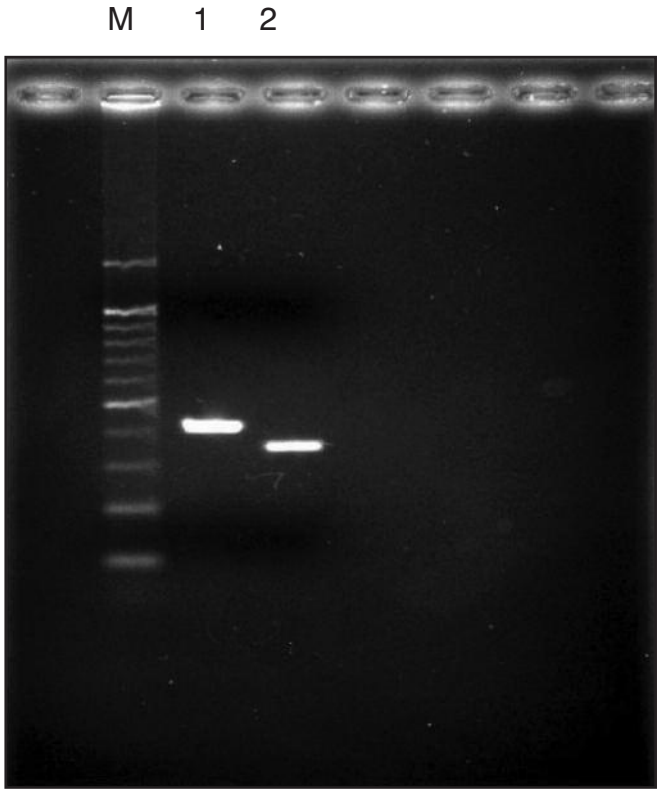

Fig. 1: Agarose gel electrophoresis of HBoV PCR product.
Line M: Marker, Line 1: using confirmation primer set (420 bp)
Line 2: using screening primer set (354 bp)

CTCTGTGTTGACTGAATACAG-3′ generating a product of 354 bp long[1]. To confirm the result, a second set of primer was employed: 5′-GACCTCTGTAAGTACTATTAC-3′ and 5′-CTCTGTGTTGACTGAATACAG-3′ with a 420-bp long product[3]. No bacterial pathogens were detected in the blood cultures.

DISCUSSION

We have successfully detected the presence of the viral genome from the patient's respiratory sample. Even though, HBoV infections are being reported worldwide since its discovery in 2005 with a range of 1.5%-33% of respiratory samples taken from children with ARTI; this is the first confirmed case of HBoV detection in Malaysia[2]. The role of HBoV as a pathogen or passenger has been subjected to debates. One of the facts that raise its suspicion as a pathogen is due to its asymptomatic persistence in the respiratory tract. Besides, it is commonly detected in association with other respiratory viruses. Furthermore, it does not belong to any family of respiratory pathogens.

Confirming a causal relationship between HBoV and symptoms is difficult[1]. Its establishment as a causative agent is complicated by the fact that it cannot be propagated in any culture systems nor there are in any animal models. The identification of the virus is mainly based on the detection of the virus DNA in the clinical samples of respiratory, blood, urine and stool using PCR techniques and further sequencing of the amplicones, as in our case. Quantification method especially detecting the virus with high viral load in blood rather than respiratory secretion shows better association with clinical symptoms[2]. However, the clinical significance of low to moderate viral loads is uncertain and may be related to asymptomatic shedding and viral persistence. As for this patient, quantification by PCR was unfortunately, not part of this study, nor was blood analysed to detect the virus. However, in the absence of other common pathogens, HBoV could be the likely pathogen.

In contrast to RSV infections, which occurred mostly in children less than 6 months, the majority of the HBoV infections were observed between 6 months and 3 years of age, in agreement with our case[4]. Fever, cough, rhinitis, sore throat, and respiratory distress, are the main symptoms of HBoV infected children[3]. It seemed that the clinical features associated with HBoV infection is no different than that of caused by respiratory viruses. These include upper RTI, bronchitis, bronchiolitis, pneumonia and acute exacerbation of asthma.

However, there is an association between acute wheezing and HBoV detection in some studies[1]. Allander found that one fifth of children with acute wheezing were attributed to HBoV infection, of which high viral load of the implicated virus was noted in the absence of other viruses, suggesting a causative role for acute wheezing[2]. HBoV pathogenecity may also be facilitated by other predisposing conditions such as asthma, or previous bronchiolitis episodes as well as history of prematurity as reported in a study on a population of 589 French children with respiratory infections, as seen in our case[5].

Here, we described the first finding of HBoV in a Malaysian child with pneumonia, and a pre-existing history of asthma. Numerous reports had indicated of its worldwide endemicity. Full epidemiological study on its prevalence and associated demographic and clinical features is ongoing and this will hopefully shed light on the seasonality and prevalence rate among children with acute lower respiratory tract infections in the country.

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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. Proceedings of the National Academy of Sciences of the United States of America. 2005; 102(36): 12891.
2. Allander T, Jartti T, Gupta S, Niesters HGM, Lehtinen P. Human bocavirus and acute wheezing in children. Clinical Infectious Diseases. 2007; 44(7): 904.
3. Bastien N, Brandt K, Dust K, Ward D, Li Y. Human bocavirus infection, Canada. 2006; 12(5).
4. Weissbrich B, Neske F, Schubert J, *et al.* Frequent detection of bocavirus DNA in German children with respiratory tract infections. BMC Infectious Diseases. 2006; 6(1): 109.
5. Foulongne V, Olejnik Y, Perez V, Elaerts S, Rodiere M, Segondy M. Human bocavirus in French children. Emerging Infectious Diseases. 2006; 12(8): 1251.