Human Bocavirus Diagnosis by Molecular Method from Respiratory Infection Patients in Mosul City

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ABSTRACT

The Bocavirus, which causes respiratory illness in the lower respiratory tract in newborns, children, and healthy adults, was investigated in this study. Methodology and results: The samples were taken as Nasopharyngeal / Throat swabs kept in sterile VTM (Viral Transport Media), which was collected from Mosul Hospitals within six months of the study. The Tested patients' ages ranged from under 20 to above 40 (between 11-57) for both sex males and females. The dominion age group was above 30, and the DNA of this virus was extracted by the real-time PCR method. Conclusion, significance, and impact of study: the molecular test indicates positive in twenty-four samples from all 70. This demonstrates the presence of Bocavirus in (20%) males and (14.28%) female understudies.

Keywords: Respiratory Tract infections, Bocavirus, Real-time PCR, Virus Transport Media, Viral Transport Media.

INTRODUCTION

The Parvovirinae and Densovirinae subfamilies, which incorporate infections that contaminate vertebrates and infections that contaminate arthropods, individually, outline the reality that parvoviridae infections contaminate a differing assortment of 1Human bocaviruses (HBoVs) are a kind of infection that has a place in the Parvoviridae family. HBoVs are minor, non-enveloped infections having icosahedral symmetry and a 5 kb single-stranded DNA genome. Two critical basic proteins, VP1 and VP2, and two non-structural proteins, NS1 and phosphoprotein 1, are encoded by the genome (NP1). Based on the nucleotide (nt) dissimilarity of the VP1 capsid locale, HBoVs are isolated into four species: HBoV1, HBoV2, HBoV3, and HBoV4 2. They have a ssDNA genome and create minor capsids. As it were, three proteins with known capacities are encoded by the viral genome: the non-structural protein NS1 and the two capsid proteins VP1 and VP2 3. Human bocavirus is found more regularly in clinical tests from patients with respiratory sicknesses. However, HBoV-2, HBoV-3, and HBoV-4 are more commonly connected to gastrointestinal diseases. However, their particular part as enteropathogens is obscure 4. Human bocavirus (HBoV) 1 was found in pediatric respiratory...
examples in 2005 and is the moment human-pathogenic parvovirus. Since then, a few inquiries have been made to see intense respiratory tract disease (ARTI) at its interface. Bronchitis, pneumonia, asthma, and the common cold are the foremost visit clinical signs of intense HBoV1 contamination (Kang et al., 2018). Human bocavirus 1-4 has been studied for its pathogenicity in patients with respiratory disease, gastroenteritis, heart disease, meningitis/encephalitis, fetal hydrops or mortality, cancer, and transplant recipients/ immunocompromised people. The core is where parvovirus replication and get-together take put, and it is subordinate to the host's cellular movement. The genome replication instrument of the viral family is interesting. The fastener structure at the 3' conclusion works as a self-primer, causing plus-sense DNA to be synthesized and double-stranded DNA to be delivered. The fastener shape is utilized as groundwork to decipher extra minus-sense strands from ds DNA. Concurring to the current demonstration, the amplified strand rehashes back on itself to create a tetrameric shape, which is along these lines partitioned into two plus-sense and two minus-sense DNA strands. Amid replication within the core, connection to have receptors triggers clathrin-mediated endocytosis of the virion into the have cells. The virion moves to the core after entering the cytoplasm. The virus's ssDNA genome enters the core. When the cell enters the S stage, cellular proteins change over ssDNA to dsDNA, and dsDNA translation occurs in viral mRNAs, which are deciphered into viral proteins. For replication, the rolling-hairpin method is utilized. The NS1 endonuclease covalently connects to the 5' genomic conclusion. These recently made ssDNA atoms can either be changed over to dsDNA and utilized as a layout for translation and replication, or they can be typified and liberated by cell lysis.

MATERIALS AND METHOD

Specimens Collection

The study that we made involved the collection of (70) samples from the nasopharyngeal and throat swabs that were taken and kept in sterile Viral Transport Media (VTM) from respiratory tract infection patients from Mosul hospitals within six months of the study. We collected the samples from Three hospitals in Mosul City (Al-Shifaa Hospital for Infectious Diseases, Al-Salam Teaching Hospital and Advisory Clinic for Chest and Respiratory Diseases at Al-Faisalia in Mosul City). The samples were immediately transported to the laboratory and kept in a deep freeze (-70°C) until they were used in viral DNA extraction.

Extraction of Viral DNA & Real-Time PCR Process

The QIAGEN QIAamp®Viral DNA Min Kit extracted viral DNA from throat swab specimens according to the manufacturer's recommendations (Germany). Polymerase chain reaction techniques have advanced much more since the development of quantitative RT-PCR. To amplify viral DNA, real-time methods are used (Fast-track Respiratory Pathogens 33 kits from Luxembourg). The FTD test was the only means to identify Bocavirus from the rest of the Bocaparvovirus family because the majority of Bocaparvovirus identified was Bocavirus. The presence of specific pathogen sequences in the process indicates a spike in fluorescence from the related dual-labeled probe, which the Real-Time thermocycler displays as a cycle threshold value (Ct). During the lysis buffer step of the extraction process, the equine arteritis virus (EAV) was used as an internal control (IC) and a negative control utilizing an Applied Biosystem Real-time PCR7500, Germany.
RESULTS
The mean age of Bocavirus respiratory tract infection in patients for all males and females were (32.16±11.82, 31.28±12.56, and 33.4±11.23) respectively. Twenty-four swabs (34.28%) out of seventy samples tested positive for Bocavirus infection in RT-PCR (Table 1). The amplification reaction appears to be Bocavirus-specific at a high CT value (30.56), while the mean CT value of Bocavirus infection for all males and females was (34.00±2.20, 34.05±2.17, 33.94±2.37) respectively (Figure 4).

Figure 4. The quantity of fluorescence acquired during each amplification cycle was shown on an amplification plot. Bocavirus was represented by the number Ct (30.56).

<table>
<thead>
<tr>
<th>AGE / Year</th>
<th>Bocavirus</th>
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<tbody>
<tr>
<td></td>
<td>M</td>
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<tr>
<td>Under 20</td>
<td>3</td>
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<tr>
<td>20 – 29</td>
<td>7</td>
</tr>
<tr>
<td>30 – 39</td>
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<tr>
<td>Above 40</td>
<td>5</td>
</tr>
<tr>
<td>Total</td>
<td>24</td>
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<td>%</td>
<td>20.00</td>
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Table 1. Shows the age and gender of patients with Bocavirus infection

DISCUSSION
Our Real-Time PCR investigation yielded a good result at (high Ct 30.56). We took seventy throat and nasopharyngeal swab samples, and twenty-four (34.28%) were positive for HBOV, which indicates the dominant age was (30 and above) for male and female patients from Mosul city in Iraq, while in contrast, other research results in Baghdad in the same country of Iraq, which indicates the more patients who infect with bocavirus been (under20) from infants and children. RSV (respiratory syncytial virus) worsens HBoV infection, even though HBoV infection has distinct epidemiological features (Huiming et al., 2018). When it comes to young children who have respiratory problems, the human bocavirus (HBoV) is commonly found. An immunocompromised child with disseminated HBoV
infection might cause life-threatening hypovolemic shock due to diarrhea. Human bocavirus infection rates in Iraq were compared to infection rates in adjacent countries. No significant differences were noticed between infection rates and different parameters except with nasal discharge and wheezing. Asymptomatic individuals with prolonged HBoV shedding and HBoV detections may be linked to HBoV persistence, at least in a yet-to-be-defined subset of patients, whereas symptomatic individuals had initial infection or reinfection. Rhinovirus, bocavirus, and adenovirus cases did not follow a seasonal pattern; they were seen all year. On the other hand, the influenza A and B viruses were present in the winter and spring. At the same time, metapneumovirus was found in the fall, winter, and spring but not in the summer. Cigarette smoke exposure was thought to enhance epithelial susceptibility to viral infection by raising the quantity of their receptor in youngsters whose relatives smoked. Cigarette smoke significantly contributes to viral respiratory tract infection, increasing morbidity and mortality and initiating latent infection. Without generating respiratory symptoms, HBoV DNA has been found in lung and colorectal tumors, showing that the virus may persist in some organs. Given that another parvovirus, B19, can survive in infected hosts, HBoV persistence is a possibility and could cause long-term illness in some patients. The human bocavirus causes acute respiratory infections (ARI) in children. It was discovered in Sweden and Australia and is thought to be responsible for 5-15 percent of all ARI cases in children (Hart and Cuevas, 2007). Cough was the most prevalent symptom across all illnesses in patients with respiratory viruses. Patients with bocavirus and parainfluenza 2–4 had the longest duration of symptoms (7 days). Women with bocavirus had the highest severity score of 22, which ranged from 6 to 50.

CONCLUSION

Bocavirus causes respiratory illness in the lower respiratory tract in newborns, children, and healthy adults. The Real-Time PCR method was the best method for diagnosis. Our study highlights RT-PCR in the diagnosis of respiratory infection. Acknowledgments: The authors are very grateful to the University of Mosul / College of Science / Biology department for their provided facilitates, which helped to improve the quality of this work. We thank the staff from Al-Shifaa Hospital for Infectious Diseases, Al-Salam Teaching Hospital, and Advisory Clinic for Chest and Respiratory Diseases at Al-Faisalia in Mosul City and Central Public Health Laboratory, Virology Department, Baghdad, Iraq, Who gave insight and experience that supported the study greatly.

References


