Scholars Journal of Applied Medical Sciences
Abbreviated Key Title: Sch J App Med Sci
ISSN 2347-954X (Print) | ISSN 2320-6691 (Online)
Journal homepage: https://saspublishers.com

OPEN ACCESS

Pathology

Molecular Characterization of Human Adenoviruses Associated with Pediatric Respiratory Infections in Omdurman, Sudan

Abdulaziz Abdirizak Saed[1], Dr. Mohammed Eldai Hamad[2], Dr. Rashid Awad Abdalla Salih[3], Dr. Mohammed Ahmed Abd Allah[4*]

[1]Post Graduate, Garb El-Neil College, Sudan
[2]Department of Microbiology, Faculty of Medical Laboratory Sciences, Dar Eloum College for Science and Technology, Khartoum, Sudan
[3]King Fahad Military Medical Complex Dhahran, KSA
[4]Department of Pathology, College of Medicine, Karary University Sudan, P.O. Box: 12304, Khartoum, Sudan

DOI: https://doi.org/10.36347/sjams.2024.v12i09.017 | **Received:** 17.08.2024 | **Accepted:** 21.09.2024 | **Published:** 25.09.2024

***Corresponding author:** Dr. Mohammed Ahmed Abd Allah
Department of Pathology, College of Medicine, Karary University Sudan, P.O. Box: 12304, Khartoum, Sudan

Abstract **Original Research Article**

Acute respiratory viral infection (ARVI) is a common cause of morbidity and mortality especially in pediatric patients. Among important respiratory pathogens are adenoviruses, The illnesses can range from the common cold to pneumonia Depending on the type, adenoviruses can cause other illnesses such as gastroenteritis, conjunctivitis, cystitis, and, less commonly, neurological disease. Aim of study: molecular detection of respiratory infection due to adenovirus among children in Khartoum state. Respiratory specimens were collected from children's patients with RTIs in the emergency department Mohamed Alamin Hamid pediatric hospital Omdurman in Khartoum state during the period of October 2022 to February 2023.the HAdV infection were determined by PCR technique. A total of 50 nasal swap were investigated for HAdV, throughout th PCR technique by using specific primer. The results showed 18 (36.0%) were positive and 32 (64.0%) were negative for HAdVs. The statistical analysis revealed no significant difference between age groups (X2= 0.786 P. value: 0.375). This study summarized the HAdV were the predominant types identified in children less than five years intend to have respiratory tract infection according to clinical symptoms of upper or lower respiratory infections.by using conventional PCR for identify with highly sensitively.

Keywords: Adenovirus; Infections; Respiratory Tract Infection; Pediatrics; Sudan.

1.1 INTRODUCTION

Acute respiratory tract infections (ARTI) are regarded as important causes of morbidity and mortality in pediatric patients. This refers to both outpatient visits and 20- 40% of hospitalized patients [1, 2]. The significant impact of these infections on developing countries is based on the fact that nearly 50% of pediatric consultations are related to the respiratory tract infection (RTI) [3]. The world health organization (WHO) ranks the lower RTIs as the leading cause of disease worldwide, which accounts for 4 million deaths per year [2, 4]. Most of these infections are caused by respiratory syncytial virus (RSV), influenza virus A or B (FluV), parainfluenza virus (PIV), rhinovirus (RV), and human adenovirus (HAdV). Several recently discovered viruses such as human meta pneumovirus (HMPV), human bocavirus (HBoV), and human corona viruses (HCoV) have been identified as potential respiratory pathogens [5].

The name (HAdVs) derives from their initial isolation from human in 1953 [6]. It infect a broad range of vertebrate hosts; in humans, more than 50 distinct adenoviral have been found to cause a wide range of illnesses, from mild respiratory infections in young children (known as the common cold) to life threatening multi-organ disease in people with a Weakened immune system [7].

This pathogen is classified among the Adenoviridae family and belongs to the genus Mastadenovirus It is a ubiquitous non enveloped virus of medium-sized double-stranded-DNA ranging from 34 kb to more than 37 kb, which encodes around 40 genes [8]. Human adenoviruses (HAdVs) are a common cause of respiratory infection in persons of all ages. Acute upper and lower respiratory tract diseases, including pneumonia and bronchitis, have been attributed to HAdVs. Although many infections are mild, some

persons, such as very young children, elderly or immunocompromised persons [9].

Human adenoviruses also (HAdV) are a major cause of clinical infections including gastroenteritis, conjunctivitis, ocular, respiratory diseases, hemorrhagic cystitis and chronic systemic infections [10]. Human adenoviruses (HAdVs) are classified into 7 species (Human mastadenovirus A to G) and at least 69 recognized genotypes based on serology, whole-genome sequencing, and phylogenetic analyses [11]. Adenovirus accounts for 5-15% of upper and lower RTIs in infants and children hospitalized for respiratory disease [2, 12]. The detection of viral nucleic acids by amplification assays [12].

2. Objectives

According to the importance of this virus, we aimed to detect of respiratory infection caused by adenovirus among children in Mohammed Alameen Hamid pediatric Hospital in Khartoum State, and show prevalence of adenovirus infection among different age group.

3. METHODOLOGY

3.1. Clinical Specimens

The specimens used in this study were collected, from throat swabs from 50 pediatric patients with RTI admitted to the MAHPH, from August 2022 to February 2023. The relevant clinical information was obtained through a standard questionnaire, containing, hospitalization status, age, sex, and clinical symptoms such as fever, cough, sneeze, and muscle ache. Throat swabs immersed in phosphate-buffered saline (PBS pH 7.2) and stored at -70℃ for subsequent DNA extraction.

3.2 Detection and isolation of adenoviruses:

DNA was done extracted by steps: cell lysis, protein precipitation and DNA precipitation ZR Viral DNA kit (Viral DNA Extraction) Epigenetics company (USA) as described by the manufacturer's instructions.

Primers

Primer sequence was used for the amplification of human adenovirus [48]. The more details in the appendix (5′-CTGATGTACTACAACAGCACTGGCAACATGGG -′3) (5′-GCGTTGCGGTGGTTAAATGGGTTTACGAT -′3). Master Mix kits containing all reagents for PCR except water and primers will be using although Storage of the master mix will be under (-20).

Amplification Conditions of PCR

The amplification was done by using (0.25) PCR Eppendorf tubes using thermocycler PCR. (SensoQuest, Germany) The amplification conditions are: denaturation annealing and extension. The pcr mixture was subjected to initial denaturation step at 94c for 5 min followed by 35 cycles of denaturation at 94c for 1 min, primer annealing at 52c for 1 min followed by step of extension at 72c for 1 min and final elongation at 72c for 10 min.

Gel Electrophoresis

Amount of (1.5 gm) of agarose powder (appendix 1) was weighted, (100 ml) 1X TBE buffer be added, the mixture was heated by microwave until clear solution is produced, allowed to cool, then (2µl) of Ethidium bromides was added, mixed well and poured on to suitable gel tray that was equipped with suitable combs to form well that used for loading the PCR products. Any bubbles was removed and the gel was allowed to solidify at room temperature. After solidification, the comb will be gently removed and the spacer from the opened sides was removed. The gel was removed by gel holder and visualized under the ultraviolet Transilluminator to detect the specific amplification sequence 100.1000pb.

3.3 Data Analysis

The collected data done in computerized and analyzed by statistical package for social sciences (SPSS).

4. RESULTS

A total of numbers of 50 nasal swabs were selected as a study group from different children less than 5 years, for the detection of HAdV DNA. Results showed that 18 (36.0%) were positive and 32 (64.0%) were negative for HAdVs as tested by a conventional PCR. The distribution of the results shows (Table 1).

The study shows the gender distribution between males (32.0%) and females (68.0%) the study shows (Table 2). And according to age of children that target in academic study level are 1 years 3(6.0%) and 2 year is 10 (20.0%) were 3 years is 18 (36.0%) And 4 years is 12 (24.0%) and 5 years is 7(14.0%) the distribution (show table 3) were the age group of children, are classified into two groups. The group one is ≤ 2 years 13 (26.0%) were s group two is > 2 years 37 (74.0%).

Regarding to respiratory tract infection where classified base on the symptoms that suffering of the patient. The upper respiratory tract infection is 30 (60.0%) were the lower respiratory tract infection is 20.

According to association of the results with age are, 1 year were positive is 1 (2%) and negative is 2 (4%) were the total is 3 (6%) and the 2 year were positive 5 (10%.0) and negative 5 (10%) were the total is 10(20%.0) and the 3 years the positive is 3 (6.0%) and negative is 15 (30.0%) were the total is 18 (36.0%). and the 4 years the positive is 5 (5.0%) and negative is 7 (14.0%) where the total is 12 (24.0%) and the 5 years the positive is 4 (8.0%) and negative is 3 (6.0%) where the total is 7 (14.0). the total result of positive is 18 (36.0%)

and the total result of negative is 32 (64.0%) where the total count is 50 (100%) the Chi-square:5.305 and P. value: 0.257. This is insignificant value of probability (P. value) according to level of (0.257) (Table 3).

According to association of the results with age group we classified into two groups The result of group one ≤ 2 years positive is 6 (12.0%) and negative is 7 (14.0%) in total of group one is 13 (26.0%) where the result of group > 2 years positive is 12 (24.0%) and negative is 25 (50.0%) in total of group two is 37 (74.0%) the association of total result count positive is18 (36.0%) and negative is 32 (64.0%) where the total count 50 (100.0%).

The Chi-square:0.786 and P. value: 0.375. This is insignificant value of probability (P. value) according to level of 0.375 (Table 4).

According to the gender HAdVs infections, this study showed no significant difference of infections between males positive 5 (10%) and negative 11 (22.0%) in total 16 (32.0%) where the females positive is 13 (26.0%) and negative 21 (42.0%) in total 34 (68.0%) the total count is 50 (100.0%). the Chi-square:0.230 and P. value: 0.631. This is insignificant value of probability (P. value) according to level of 0.631 (Table 5).

Regarding to the association symptoms of respiratory tract infection the academic level show no significant difference correlation. The result of upper respiratory tract infection positive is 12 (24.0%) and negative is 18 (36.0%) in total 30 (60.0%) where the result of lower respiratory tract infection positive is 6 (12.0%) and negative is 14 (28.0%) in total 20 (40.0%) the total count is 50 (100.0%). the Chi-square:0.512 and P. value: 0.470. This is insignificant value of probability (P. value) according to level of 0.470 (Table 6).

Distribution of Results

Table 1: Frequency of Negative and Positive Results of Adenovirus Samples

Result	Frequency	Percent
Positive	18	36.0
Negative	32	64.0
Total	50	100.0

Distribution of Gender

Table 2: Frequency of Distribution of gender

Gender	Male	16	32.0
	Female	34	68.0
	Total	50	100.0

Table 3: Association of results with age

Age		Result		Total
		Positive	Negative	
1	Count	1	2	3
	%	2.0%	4.0%	6.0%
2	Count	5	5	10
	%	10.0%	10.0%	20.0%
3	Count	3	15	18
	%	6.0%	30.0%	36.0%
4	Count	5	7	12
	%	10.0%	14.0%	24.0%
5	Count	4	3	7
	%	8.0%	6.0%	14.0%
Total	Count	18	32	50
	%	36.0%	64.0%	100.0%
Chi-Square:5.305 P.value: 0.257				

Table 4: Association of results with age group

Age group		Result		Total
		Positive	Negative	
≤ 2 years	Count	6	7	13
	%	12.0%	14.0%	26.0%
>2 years	Count	12	25	37
	%	24.0%	50.0%	74.0%
Total	Count	18	32	50
	%	36.0%	64.0%	100.0%
Chi-Square:0.786 P.value: 0.375				

Table 5: Association of Results with Gender

Gender		Result		Total
		Positive	Negative	
Male	Count	5	11	16
	%	10.0%	22.0%	32.0%
Female	Count	13	21	34
	%	26.0%	42.0%	68.0%
Total	Count	18	32	50
	%	36.0%	64.0%	100.0%
Chi-Square:0.230 P.value: 0.631				

Table 6: Association of Results with Respiratory

Respiratory		Result		Total
		Positive	Negative	
Upper	Count	12	18	30
	%	24.0%	36.0%	60.0%
Lower	Count	6	14	20
	%	12.0%	28.0%	40.0%
Total	Count	18	32	50
	%	36.0%	64.0%	100.0%
Chi-Square:0.512 P.value: 0.470				

5. DISCUSSION

Respiratory infection is a leading cause of morbidity, hospitalization, and death in pediatric patients [12]. Adenoviruses are most important respiratory pathogens, and constitute 5-10% of all RTIs in infants and children younger than two years of age and 1–7% in adults, in patients with severe HAdV pneumonia, the mortality rate may exceed 50% [12].

Adenovirus infections can be diagnosed by a variety of traditional and molecular methods. Molecular methods are most sensitive for detecting adenovirus in clinical specimens. The present study was conducted to detect HAdV by nested PCR. The frequency of HAdV in our study was 36% (18 out of 50 samples) that is slightly higher than the frequency of 22% reported by another study from Iran [13, 14], and several parts of the world including Oman (15%), Kenya (14%), Brazil (10%), Korea (10.3%), Australia (7.3%), Hung Kong (5.3%), Mejia (5.2%), and China (4.9%) [15-17]. This high results in our study due to small size of the study (50 samples). In the present study According to the age group of children, were classified into two groups.the group is ≤ 2 years 13 (26.0%) were other group is > 2 years 37 (74.0%) where finding there no difference correlation between them and the statistical result show insignificant of P .value is 0.786 these finding agree with study done by Moattari *et al*., [18]. that stated the P. value is 0.527. but This study finding disagree with significant result p. value 0.01. Khalaf R, Fadhil H *et al*., [19].

According to the gender this study showed no significant difference in HAdVs infections between males 16 (32.0%) and females 34 (68.0%) the P. value is 0.230. the other study agree with no significant of P. value is 0.299. also showed between males and females [21, 19]. But this study disagrees with J.S. Ampuero *et al*., by significant p. value 0.017 [22]. Adenovirus plays a role respiratory tract infection, regarding to the study show there no significant correlation between upper respiratory tract infection and lower respiratory tract infection the P. value is 0.512. But these finding disagree with study done by Moattari *et al*., [18]. 0.003 0.004.4.2.

6. CONCLUSION

This study provides the molecular in a total of 50 sample of nasal swap, were identified human adenovirus infection in children less than five years with suffering in respiratory tract infection and This study provides the infection were those have clinical symptoms of upper or lower respiratory tract infection the study concluded was insignificant and No correlation observed in respiratory of human adenovirus infection with age and gender.

REFERENCE

1. Kalu, S. U., Loeffelholz, M., Beck, E., Patel, J. A., Revai, K., Fan, J., ... & Chonmaitree, T. (2010). Persistence of adenovirus nucleic acids in nasopharyngeal secretions: a diagnostic conundrum. *The Pediatric infectious disease journal*, *29*(8), 746-750.
2. Marinheiro, J. C., Sanalios, R. B., Santos, D. C. D., Costa, C. A. D., & Hársi, C. M. (2009). Duplex-PCR assay for the detection of adenovirus and respiratory syncytial virus in nasopharyngeal samples. *Memórias do Instituto Oswaldo Cruz*, *104*, 118-120.
3. Abd-Jamil, J., Teoh, B. T., Hassan, E. H., Roslan, N., & AbuBakar, S. (2010). Molecular identification of adenovirus causing respiratory tract infection in pediatric patients at the University of Malaya Medical Center. *BMC pediatrics*, *10*, 1-7.
4. Dominguez, S. R., Robinson, C. C., & Holmes, K. V. (2009). Detection of four human coronaviruses in respiratory infections in children: a one-year study in Colorado. *Journal of medical virology*, *81*(9), 1597-1604.
5. Albuquerque, M. C. M., Pena, G. P., Varella, R. B., Gallucci, G., Erdman, D., & Santos, N. (2009). Novel respiratory virus infections in children, Brazil. *Emerging infectious diseases*, *15*(5), 806-808.
6. Rowe, W. P., Huebner, R. J., Gilmore, L. K., Parrott, R. H., & Ward, T. G. (1953). Isolation of a cytopathogenic agent from human adenoids undergoing spontaneous degeneration in tissue culture. *Proceedings of the Society for Experimental Biology and Medicine*, *84*(3), 570-573.
7. H: Double-Stranded DNA Viruses- Adenoviruses". *Biology Libre Texts*. 2017 25 June. Retrieved 6 January 2021.
8. Black, R. E., Cousens, S., Johnson, H. L., Lawn, J. E., Rudan, I., Bassani, D. G., ... & Mathers, C. (2010). Global, regional, and national causes of child mortality in 2008: a systematic analysis. *The lancet*, *375*(9730), 1969-1987.
9. Ison, M. G. (2006). Adenovirus infections in transplant recipients. *Clin Infect Dis*, 43, 331-339. doi: 10.1086/505498
10. De Jong, J. P., Bruins, A., Dolfsma, W., & Meijaard, J. (2003). Innovation in service firms explored: what, how and why.
11. Refrence Harrach, B., Benkő, M., Both, G. W., Brown, M., Davison, A. J., & Echavarría, M. (2011). Family Adenoviridae. In: King, A. M. Q., Adams, M. J., Carstens, E. B., Lefkowitz, E. J., editors. Virus Taxonomy. 9th Report of the International Committee on Taxonomy of Viruses. New York: Elsevier; pp. 125–141.
12. Shike, H., Shimizu, C., Kanegaye, J., Foley, J. L., & Burns, J. C. (2005). Quantitation of adenovirus genome during acute infection in normal children. *The Pediatric infectious disease journal*, *24*(1), 29-33.
13. Ward, T. G. (December 1953). Isolation of a cytopathogenic agent from human adenoids undergoing spontaneous degeneration in tissue culture. *Proceedings of the Society for Experimental Biology and Medicine*, 84(3), 570-573.
14. Ou, Z. Y., Zeng, Q. Y., Wang, F. H., Xia, H. M., Lu, J. P., Xia, J. Q., ... & Zhou, R. (2008). Retrospective study of adenovirus in autopsied pulmonary tissue of pediatric fatal pneumonia in South China. *BMC infectious diseases*, *8*, 1-6.
15. Yeung, R., Eshaghi, A., Lombos, E., Blair, J., Mazzulli, T., Burton, L., & Drews, S. J. (2009). Characterization of culture-positive adenovirus serotypes from respiratory specimens in Toronto, Ontario, Canada: September 2007–June 2008. *Virology journal*, *6*(11), 1-3.
16. Zhang, H. Y., Li, Z. M., Zhang, G. L., Diao, T. T., Cao, C. X., & Sun, H. Q. (2009). Respiratory viruses in hospitalized children with acute lower respiratory tract infections in harbin, China. *Japanese journal of infectious diseases*, *62*(6), 458-460.
17. Moattari, A., Emami, A., Pirbonyeh, N., & Yaghoobi, R. (2014). Detection of Adenovirus Infection among Children with A cut Respiratory Disease during in Shiraz, Iran (2010-2012). *Arch pediatr Infect Dis*, *2*(4), 5930-5945.
18. Khalaf, R. J., Fadhil, H. Y., Aufi, I. M., & Namdar, S. A. (2017). Molecular diagnosis of human adenovirus in children with upper respiratory tract infections. *Journal of Pharmacy and Biological Sciences*, *12*(3), 09-13.
19. Zou, L., Zhou, J., Li, H., Wu, J., Mo, Y., Chen, Q., ... & Ke, C. (2012). Human adenovirus infection in children with acute respiratory tract disease in G uangzhou, C hina. *Apmis*, *120*(8), 683-688.
20. Mitchell, L. S., Taylor, B., Reimels, W., Barrett, F. F., & Devincenzo, J. P. (2000). Adenovirus 7a: a community-acquired outbreak in a children's hospital. *The Pediatric infectious disease journal*, *19*(10), 996-1000.
21. Ampuero, J. S., Ocaña, V., Gómez, J., Gamero, M. E., Garcia, J., Halsey, E. S., & Laguna-Torres, V. A. (2012) Adenovirus Respiratory Tract Infection in Peru. PLoS One, 7, e46898
22. Adenovirus Clinical Overview for Healthcare Professionals CDC". *www.cdc.gov*. 29 November 2021. Retrieved 5 May 2022.

23. Lu, X., & Erdman, D. D. (2006). Molecular typing of human adenoviruses by PCR and sequencing of a partial region of the hexon gene. *Arch Virol*, *151*(8), 1587-1602.
24. Brealey, J. C., Sly, P. D., Young, P. R., & Chappell, K. J. (2015). Viral bacterial co-infection of the respiratory tract during early childhood. *FEMS microbiology letters*, *362*(10), fnv062.
25. Miceli, I. Ministerio de Salud Pública y Acción Social, personal Communication.
26. Sun, Q., Jiang, W., Chen, Z., Huang, L., Wang, Y., Huang, F., ... & Yan, Y. (2014). Epidemiology and clinical features of respiratory adenoviral infections in children. *European journal of pediatrics*, *173*, 441-444.
27. Henrickson, K. J. (2004). Advances in the laboratory diagnosis of viral respiratory disease. *The Pediatric infectious disease journal*, *23*(1), S6-S10.
28. Garbino, J., Gerbase, M. W., Wunderli, W., Deffernez, C., Thomas, Y., Rochat, T., ... & Kaiser, L. (2004). Lower respiratory viral illnesses: improved diagnosis by molecular methods and clinical impact. *American journal of respiratory and critical care medicine*, *170*(11), 1197-1203.
29. Tesini, B. L. (April 2022). *MSD Manual Professional Edition*. Retrieved 7 May 2022.
30. Ephros, M., Friedman, B. C., Elhasid, R., Kra-Oz, Z., Shaked-Mishan, P., Sattinger, J., & Kassis, I. (2009). Incidence and clinical manifestations of adenoviral infection among children undergoing allogeneic stem cell transplantation. *Isr Med Assoc J*, *11*(12), 744-748.
31. Sarantis, H., Johnson, G., Brown, M., Petric, M., & Tellier, R. (2004). Comprehensive detection and serotyping of human adenoviruses by PCR and sequencing. *Journal of Clinical Microbiology*, *42*(9), 3963-3969.
32. Lynch, J. P. III., Fishbein, M., & Echavarria, M. (2011). Adenovirus. *Semin Respir Crit Care Med*, 32(4), 494–511.
33. Anderson, R. M., May, R. M. (1991). *Infectious Diseases of Humans. Dynamics and Control.* Oxford University Press; Oxford.
34. Benenson, A. S., editor. (1995). *Control of Communicable Diseases in Man.* 16th edn. American Public Health Association; Washington, DC.
35. Singh-Naz, N., Brown, M., & Ganeshananthan, M. (1993). Nosocomial adenovirus infection: molecular epidemiology of an outbreak. *Pediatr Infect Dis J*, 12(11), 922-925.
36. Hllleman, M. R., & Werner, J. H. (1954). Recovery of new agent from patients with acute respiratory illness. *Proceedings of the Society for Experimental Biology and Medicine*, *85*(1), 183-188.
37. Houng, H. S. H., Gong, H., Kajon, A. E., Jones, M. S., Kuschner, R. A., Lyons, A., ... & Metzgar, D. (2010). Genome sequences of human adenovirus 14 isolates from mild respiratory cases and a fatal pneumonia, isolated during 2006-2007 epidemics in North America. *Respiratory research*, *11*, 1-8. 10.1186/1465-9921-11-116
38. Neumann, R., Genersch, E., & Eggers, H. J. (1987). Detection of adenovirus nucleic acid sequences in human tonsils in the absence of infectious virus. *Virus research*, *7*(1), 93-97.
39. Ryu, W. S. (2017). Virus life cycle. *Molecular virology of human pathogenic viruses*, 31-45. Published online 2016 May 6. doi: 10.1016/B978-0-12-800838-6.00003-5
40. Arnberg, N. (2012). Adenovirus receptors: Implications for targeting of viral vectors. *Trends Pharmacol Sci*, 33, 442–448.
41. Wickham, T. J., Mathias, P., Cheresh, D. A., & Nemerow, G. R. (1993). Integrins αvβ3 and αvβ5 promote adenovirus internalization but not virus attachment. Cell, 73, 309-319.
42. Lynch, J. P., & Kajon, A. F. (2016). Adenovirus: Epidemiology, Global Spread of Novel Serotypes, and Advances in Treatment and Prevention. *Semin Respir Crit Care Med*, *37*, 586–602.
43. Khanal, S., Ghimire, P., & Dhamoon, A. S. (2018). The Repertoire of Adenovirus in Human Disease: The Innocuous to the Deadly. *Biomedicines 6*, 30.
44. Allard, A., Albinsson, B. O., & Wadell, G. (2001). Rapid typing of human adenoviruses by a general PCR combined with restriction endonuclease analysis. *Journal of clinical microbiology*, *39*(2), 498-505.
45. Flomenberg, P., Gutierrez, E., Piaskowski, V., & Casper, J. T. (1997). Detection of adenovirus DNA in peripheral blood mononuclear cells by polymerase chain reaction assay. *J Med Virol*, 51, 182-188.
46. Wilder-Smith A., & Freedman, D. O. (2020). Isolation, quarantine, social distancing and community containment: pivotal role for old-style public health measures in the novel coronavirus (2019- nCoV) outbreak. *Journal of Travel Medicine*, 13.
47. Responding to community spread of COVID-19 [online] Website https://www.who.int/ publications-detail/responding-to-community-spread-ofcovid-19. 2020.
48. Using face masks in the community reducing COVID-19 transmission from potentially asymptomatic or pre-symptomatic people through the use of face masks or presymptomatic people through the use of face masks ECDC Technical Report [online]. Website https://www.ecdc.europa.eu/en/publications-data/using-face-masks-community reducing- covid-19-transmission. 2020
49. Brenda, L. T. (2022). University of Rochester School of Medicine and Dentistry Last review/revision.
50. Barrett B., Locken K., Maberry R., Schwamman J., Bobula J., & Brown R. (2002) The wisconsin upper respiratory symptom survey: development of an

instrument to measure the common cold. *J Fam Pract*, *51*(3), 265–273.
51. Barrett, B., Brown, R. L., Mundt, M. P., Safdar, N., Dye, L., & Maberry, R. (2005). The wisconsin upper respiratory symptom survey is responsive, reliable, and valid. *J Clin Epidemiol*, 58, 609-617.
52. Allan, G. M., & Arroll, B. (2014). Prevention and treatment of the common cold: making sense of the evidence. *CMAJ*, *186*(3), 190–199.
53. Brenda, L. T. University of Rochester School of Medicine and Dentistry.
54. Liu, C., Xiao, Y., Zhang, J., Ren, L., Li, J., Xie, Z., ... & Shen, K. (2015). Adenovirus infection in children with acute lower respiratory tract infections in Beijing, China, 2007 to 2012. *BMC infectious diseases*, *15*(1), 1-9.
55. Brini, I., Guerrero, A., Ezzine, I. K., Orth-Höller, D., Hetzer, B., Würzner, R., ... & Stoiber, H. (2020). Human adenoviruses associated with respiratory illness in neonates, infants, and children in the Sousse area of Tunisia. *Journal of Medical Virology*, *92*(12), 3081-3092.
56. Carballal, G., Videla, C., Misirlian, A., Requeijo, P., & Aguilar M. (2002). Adenovirus type 7 associated with severe and fatal acute lower respiratory infections in Argentine children. *BMC Pediatrics*, *2*(1).
57. Pierangeli, A., Gentile, M., Di Marco, P., Pagnotti, P., Scagnolari, C., Trombetti, S., ... & Antonelli, G. (2007). Detection and typing by molecular techniques of respiratory viruses in children hospitalized for acute respiratory infection in Rome, Italy. *Journal of medical virology*, *79*(4), 463-468.
58. Ebrahim, F. A., Mustafa, M. O., El Hussein, A. R. M., Elkhidir, I. M., & Enan, K. A. Molecular detection of respiratory syncytial virus genotypes in hospitalized children with bronchiolitis in Khartoum State 2015-2016.
59. Elhag, W., Saeed, H., Omer, E., & Ali, A. (2013). Prevalence of rotavirus and adenovirus associated with diarrhea among displaced communities in Khartoum, Sudan. *BMC Infectious Diseases*, *13*(1).
60. Crawford-Miksza, L., & Schnurr, D. P. (1996). Analysis of 15 adenovirus hexon proteins reveals the location and structure of seven hypervariable regions containing serotype-specific residues. *J Virol*, 70**,** 1836-1844.
61. Moattari, A., Emami, A., Pirbonyeh, N., & Yaghoobi, R. (2014). Detection of Adenovirus infection among children with acute respiratory disease during 2010-2012 in Shiraz, Iran. *Archives of Pediatric Infectious Diseases*, *2*(4), 5930.