Analysis of respiratory virus detection in hospitalized children with acute respiratory infection during the COVID-19 pandemic in East China

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Abstract

Objective During the COVID-19 pandemic, the adopted containment measures have affected other respiratory virus epidemiology. Consequently, we aimed to describe the characteristics of respiratory virus infection in hospitalized pediatric acute respiratory infections (ARIs) patients prior to and throughout COVID-19 in East China. Methods Nasopharyngeal secretions were collected from 9782 pediatric ARIs patients hospitalized in Shaoxing Maternal and Child Health Hospital from January 2018 to December 2022. Changes in positivity viral detection rates and epidemiological as well as clinical characteristics were analyzed and compared. Results 1633 strains of 7 common respiratory viruses were detected, with a total positive rate of 16.37% (821/5021) in 2018-2019 and 17.06% (812/4761) in 2020-2022. Compared with 2018-2019, RSV positive rate increased significantly in 2020-2022, while ADV, PIV-2, PIV-3, and flu-B detection rates were significantly reduced ($P < 0.05$). The RSV-positive rate in winter increased significantly more than in other seasons ($P < 0.05$), while PIV-3 was mainly prevalent in spring and summer. Conclusion During the COVID-19 pandemic, there were significant age distribution and seasonal differences in respiratory virus infection among hospitalized children with ARIs in East China.

Introduction

Acute respiratory infections (ARIs) tend to occur in childhood and seriously affect their health[1], as over 80% of respiratory tract infections in children result from viruses[2], particularly common ones include: adenovirus (ADV), respiratory syncytial virus (RSV), influenza virus (Flu) types A and B, and parainfluenza virus (PIV) type 1, 2, and 3 [3–5]. Respiratory viruses were shown to have their own epidemic characteristics and are easily affected by environmental, climate, and human flow factors[6,7]. To confront epidemic respiratory viruses, non-pharmaceutical interventions (NPIs) are frequently the first line of defense used for delaying and moderating their spread in the population [8].

When COVID-19 was declared a pandemic on January 2020, a set of public health preventive NPIs measures was adopted to control the disease transmission, including social withdrawal, closing schools, wearing masks, travel restrictions, personal hygiene improvements, and closing borders in China which eventually led to a significant decrease in other common respiratory virus circulation in nearly the entire temperate zone[9,10]. For investigating the COVID-19 effect and correlated preventive measures on common respiratory virus epidemiology, especially RSV, the viral pathogen epidemiological features were retrospectively analyzed in clinical samples obtained from ARIs children in East China from January 2018 to December 2019 and compared to those from January 2020 to December 2022, aiming to develop the preventive measures for viral infections in children.

Methods

Study design and participants

9782 ARIs children were gathered from pediatric patients aged < 14 years who were diagnosed at Shaoxing Maternal and Child Health Hospital from January 2018 to December 2022. According to the criteria of the U.S. Centers for Disease Control and Prevention, the inclusion criteria were: (1) children < 18 years; (2) verified fever with a body temperature exceeding 37.5; (3) one or more respiratory symptoms during the 14-day commencement (cough, sore throat, sputum, shortness of breath, lung auscultation abnormality (rale or wheeze), tachypnea, and chest pain)[11]. Meanwhile, the exclusion criteria were children: (1) having more than a visit during a week; (2) having a hospital infection; (3) with congenital pulmonary airway malformation and an impaired immune system; (4) being infected with COVID-19. These patients were screened for several respiratory pathogens. The study was approved by the Medical Ethics Committee of Shaoxing Maternal and Child Health Care Hospital (2021006).

Sample collection and laboratory test

Respiratory samples (throat swabs) were collected from the included ARIs patients within 24 h of hospitalization. Professional staff will rotate the nasopharyngeal swab back and forth 2 to 3 times, then quickly remove the swab and put it into the microbial test tube containing sterile normal saline. Those without phlegm will be atomized and attracted, and the specimen will be immediately sent to the laboratory two hours after collection.

RSV, ADV, Flu A and B, and PIV1, 2, and 3 were detected by direct immunofluorescence technique. The rapid test kit is provided by Shanghai Haide Diagnostic Co., LTD. (lot number: 186325). The specific detection principle and results were determined as follows: the nasopharyngeal secretions were repeatedly blown with a capillary straw and transferred to a centrifuge tube at the tip base, centrifuged at (400 ~ 600)×g for 5 ~ 10min to remove the supernatant and leave precipitation. It is used to add 1 drop of corresponding fluorescent antibody to each cell point of the specimen or photo, incubate in a 37.0 temperature box for 30min, and then add 1 drop of blocking solution after washing with washing liquid, and observe the results under a fluorescence microscope after sealing the piece. The FITC-labeled virus-specific monoclonal antibody binds to the corresponding viral antigen in the cell to form an antigen-antibody complex. Under the fluorescence microscope, the cells show apple green fluorescence and the negative film is stained red by Evans blue. If two green fluorescent cells are found in each field of vision under the microscope at 200 times, it is positive.

Statistical analysis

SPSS19.0 software was used to establish a database, reporting the categorical variables as frequencies and percentages. The chi-square test or Fisher exact probability method was utilized for the comparison between groups. p<0.05 indicated a significant difference.

Results

General clinical features

Among the 9782 children with ARIs were detected, 5021 cases were diagnosed before COVID-19 (2018-2019), and 4761 patients were diagnosed during the COVID-19 pandemic (2020 to 2022), with no significant difference in gender and age between 2018-2019 and 2020-2022 ($p > 0.05$). (Table 1)

Overall detection of respiratory viruses

One or more viruses were detected in 821/5021 (16.35%) samples before COVID-19, which exhibited a significant decrease in 812/4761 (17.06%) during COVID-19 (Table 1). The total detection rate of the respiratory virus showed a decreasing trend from January to April and an increasing trend from October to December, both before and during the outbreak (Figure.1A).

Prior to and during COVID-19, RSV was the most detected, accounting for 50.79% of positive samples in 2018-2019 and 76.48% in 2020 to 2022. Furthermore, RSV positive rate in 2018-2019 increased more significantly than in 2020 to 2022 (9.38% vs. 13.04%, $p < 0.05$). Nevertheless, ADV, PIV2 and 3, and Flu B positive rates in 2020 to 2022 reduced more significantly than in 2018 to 2019 ($p < 0.05$), while Flu A and PIV1 positive rates revealed no significant differences between 2018 to 2019 and 2020 to 2022 ($p > 0.05$) (Table 1).

As shown in Figure.1B, RSV infections and positive rates exhibited seasonal fluctuations annually between September and May from 2018 to 2019, 2020 to 2021, and 2021 to 2022, whereas they increased sharply in December 2019 to January 2020. ADV infections and positive rates had seasonal fluctuations annually between January 2019 and January 2020, whereas they dropped sharply in April 2020 to January 2022. Flu A infections and positive rates were low between April 2020 and April 2022, while they increased modestly in children during the COVID-19 recovery period from May 2022 to September 2022. Flu B infections and positive rates were low between April 2020 and April 2021, while they increased modestly after July 2021 but were still lower than in the same period before the COVID-19 pandemic. PIV1, 2, and 3 infections decreased after January 2020 (Figure.1C-1H).

Herein, 11 samples detected two viruses, from which 6 specimens were before COVID-19 and 5 during COVID-19 (0.12% vs. 0.10%,$p > 0.05$). Throughout COVID-19, ADV plus PIV3 detection was the most common mixed infection type, reaching 25.71% of mixed infection samples. However, Neither before nor during COVID-19 were any samples detected more than two viruses.

Comparison of the age distribution of respiratory viruses

Patients were assigned into four age groups. Table 2 shows respiratory virus detection rates in different age groups. Prior to and throughout COVID-19, the total positive rate reached the peak of 19.94% and 23.15%, respectively, in 1-12-month-children, which declined with the increase in the age of the enrolled children. In comparison to before COVID-19, the total positive rates in 0-12-month-children during COVID-19 were significantly higher ($p < 0.05$), while were significant differences in other age groups.

The predominant viruses vary between the age groups. Before and during COVID-19, and although the detection in each age group did not reveal all seven viruses, RSV was the most dominant in those < 7 years. In 2018-2019, the second dominant viruses in <3 years group were PIV-3, in 3-7years group was AVD. In 2020-2022, the second dominant virus in the < 1-year group was PIV-3, and in 1-7 years was Flu A. RSV detection rate in different age children was higher in 2020-2022 than in 2018-2019, especially in infants ($p < 0.001$).

The seasonal distribution of respiratory viruses

Table 3 shows each virus-positive sample detection relying on the month before and during COVID-19. Overall, prevalent respiratory viruses were detected more in winter than in other seasons, with 30.90% and 33.24% total positive rates before COVID-19, and 33.24% during COVID-19, respectively. Compared to the same period prior to COVID-19, the total positive rates in winter during COVID-19 were not a significant difference (p <0.05). In 2020-2022, the total positive rates in other seasons exhibited a significant reduction ($p < 0.05$).

Furthermore, Figure 2 shows that before and during COVID-19, RSV positive rate in winter was significantly higher than in other seasons ($p < 0.05$). PIV3 positive rate was higher (2.61%) in summer during COVID-19, but it was higher (3.05%) in winter before COVID-19. Conversely, PIV3 positive rate was lower (0.3%) in the summer during COVID-19, but it was higher (5.76%) in the summer before COVID-19.

Discussion

In January of 2020, Zhejiang province activated its Level 1 emergency reaction to the COVID-19 pandemic. Several NPIs were adopted for the purpose of suppressing or mitigating the virus spread. Respiratory tract virus infection in children has always been the focus of attention at home and abroad. Tremendous efforts were made to minimize common childhood respiratory virus prevalence apart from COVID-19 containment [2,12,13]. Herein, virus-positive sample numbers increased by 7.2% compared to before COVID-19. The detection percentages were relatively unaffected; this increase in numbers may be the result of a combination of factors, including a presumed decline in population immunity and a loosening of COVID-19 limitations.

More specifically, we found that the test positivity rate is extremely high over the period of the COVID-19 outbreak; the strongest contribution is provided by RSV, whose prevalence has experienced a significant increase, consistent with results described by reports from Australia[13,14], and New Zealand, likely corresponding to relaxing NPIs. These phenomena may be associated with the concept of "immunity debt"[15,16], emphasizing the necessity of recognizing NPIs as a double-edged sword; this highlights the criticality of active and ongoing epidemiological surveillance and timely adjustment of immunization approaches. The epidemiological trend of RSV infection in children had undergone a significant alternation before and after COVID-19 in East China. A significant reduction in the ADV, Flu B, and PIV-2 and -3 during COVID-19 was identified, which was in the same line with observations from other reports[17,18].

Herein, children between the ages of 1-12 months are the most susceptible to respiratory viruses, particularly RSV, both prior to and throughout COVID-19; surprisingly, compared to before COVID-19, respiratory infection prevalence was almost higher after COVID-19 in this age group, where it decreased significantly in other age groups. When a virus infects a host, the immune system is activated to fight the pathogenic microorganism[19]. In children under one year old, lacking complete immune memory and decreased innate and adaptive immunity due to the immature immune system may be liable to their respiratory-virus vulnerability, with NPIs having little influence[20]. Nevertheless, neonates receiving massive passive antibodies from mothers and older children whose immune systems are more thoroughly developed may benefit to a great extent from NPIs implementation in their protection against respiratory viral infections.

This study showed that some respiratory viruses had obvious seasonality, and the detection rates of RSV and Flu A were high in winter. During COVID-19, PIV-3 was detected mainly in autumn and winter, while before COVID-19, PIV-3 was detected mainly in summer, followed by spring. Since Flu is mainly transmitted by droplets and contact, the prevention and control measures taken in the early stage of the novel coronavirus outbreak were just in spring and summer, basically the same as those taken for flu prevention and control. It can effectively block the transmission of Flu. Nevertheless, at the beginning of the fall semester in September 2020, in the light of the Chinese domestic COVID-19 outbreak in remission, people generally went back to normal work and life in low-risk areas, like those prior to COVID-19; this may clarify the spike in PIV-3 infection in summer of 2020, consistent with relevant research results[21].

This study has some drawbacks as it is only a single-center study; besides conducting a retrospective analysis and collecting the data of children admitted to our hospital, hence the other factors which may relate to respiratory virus infection were not analyzed.

In conclusion, RSV infection is the most frequent respiratory tract infection, with significant differences in distribution between different ages and different seasons. COVID-19 prevention and control measures can effectively contain ADV, Flu A and B, and PIV1, 2, and 3 transmissions. Herein, the respiratory virus was detected and analyzed in hospitalized children with acute respiratory tract infection to provide evidence for the clinical diagnosis and treatment of respiratory tract virus infection during the epidemic period.

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Author's contribution

Jianwei Zhang conceived and designed the study. Liyan Mo and Ruoya Wu wrote the manuscript. Liyan Mo performed the data analysis. All of the authors have reviewed and approved the final version of the manuscript.

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Table 1 Epidemiologic characteristics of respiratory virus detection in hospitalized children with ARIs during 2018-2022

	2018-2019 (n=5021)	2020-2022 (n=4761)	X^2 value	P value
Characteristics,n(%)				
Age				
0~<1y	2688(53.53)	2311(48.54)	24.400	0.000
1~<3y	1223(24.36)	1256(26.38)	5.288	0.021
3~<7y	989(19.70)	1036(21.76)	6.335	0.012
7~<14y	121(2.41)	158(3.32)	7.284	0.007
Gender				
Male	2948(58.70)	2799(58.8)	0.006	0.939
Female	2073(41.30)	1962(41.2)		
Detection of viruses,n(%)				
ADV	36(0.72)	14(0.29)	8.678	0.003
RSV	471(9.38)	621(13.04)	33.061	0.000
Flu A	56(1.12)	54(1.13)	0.008	0.929
Flu B	35(0.70)	13(0.27)	8.998	0.003
PIV-1	33(0.66)	22(0.46)	1.665	0.197
PIV-2	31(0.62)	5(0.11)	21.674	0.000
PIV-3	159(3.17)	83(1.74)	20.520	0.000
Total	821(16.35)	812(17.06)	0.871	0.035
Mixed virus-positive specimens				
ADV+PIV-2	1(0.02)	0(0.00)	0.948	0.330
ADV+PIV-3	1(0.02)	0(0.00)	0.948	0.330
RSV+PIV-1	1(0.02)	1(0.02)	0.001	0.970
RSV+PIV-3	1(0.02)	4(0.08)	1.965	0.161
PIV-2+PIV-3	1(0.02)	0(0.00)	0.948	0.330
PIV-1+PIV-3	1(0.02)	0(0.00)	0.948	0.330
Total	6(0.12)	5(0.10)	0.046	0.831
Total	827(16.47)	817(17.16)	0.831	0.362

Table 2 Comparison age distribution of respiratory virus infection in hospitalized children with ARIs between 2018-2019 and 2020-2022

Year	Age Group(Y)	Total number of cases	Percentage of virus-positive specimens [n(%)] ADV	Percentage of viru[s-positive specimens [n(%)]] RSV
2018-2019	infant(0~<1)	2688	4(0.15)	352(13.10)
	child(1~<3)	1223	10(0.82)	76(6.21)
	preschool(3~<7)	989	22(2.22)	41(4.15)
	school age(7~<14)	121	0(0.00)	2(1.65)
	X^2		38.241	98.442
	P		0.000	0.000
Year	Age Group(Y)	Total number of cases	Percentage of virus-positive specimens [n(%)] ADV	Percentage of viru RSV
2020-2022	infant(0~<1)	2311	2(0.09)	443(19.17)
	child(1~<3)	1256	5(0.40)	119(9.47)
	preschool(3~<7)	1036	7(0.68)	58(5.60)
	school age(7~<14)	158	0(0.00)	1(0.63)
	X^2		9.470	162.647
	P		0.024	0.000

Table 3 Comparison of seasonal distribution of respiratory virus infection among hospitalized children with ARIs between 2018-2019 and 2020-2022

Time	Total number of cases	Percentage of virus-positive specimens [n(%)] ADV	Percentage RSV
Winter in 2018-2019 year(12m~2m)	1343	8(0.60)	334(24.87)
Winter in 2020-2022 year(12m~2m)	1501	6(0.40)	415(27.65)
X^2		0.556	2.821
P		0.456	0.093
Time	Total number of cases	Percentage of virus-positive specimens [n(%)] ADV	Percentage RSV
Spring in 2018-2019 year(3m~5m)	1254	4(0.32)	62(4.94)
Spring in 2020-2022 year(3m~5m)	997	3(0.30)	49(4.91)
X^2		0.006	0.001
P		0.939	0.974
Time	Total number of cases	Percentage of virus-positive specimens [n(%)] ADV	Percentage RSV
Summer in 2018-2019 year(6m~8m)	1163	18(1.55)	9(0.77)
Summer in 2020-2022 year(6m~8m)	998	3(0.30)	18(1.80)
X^2		8.681	4.616
P		0.003	0.032
Time	Total number of cases	Percentage of virus-positive specimens [n(%)] ADV	Percentage RSV
Autumn in 2018-2019 year(9m~11m)	1261	6(0.48)	66(5.23)
Autumn in 2020-2022 year(9m~11m)	1265	2(0.16)	139(10.99)
X^2		2.019	28.040
P		0.155	0.000

Figure legends

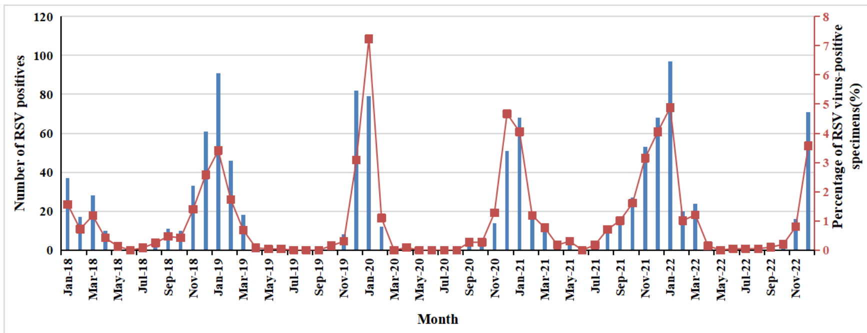
Figure 1. The number of positive and positive rates of respiratory virus infection among hospitalized children with ARIs during 2018 to 2022. A:The number of positive and positive rates of all respiratory

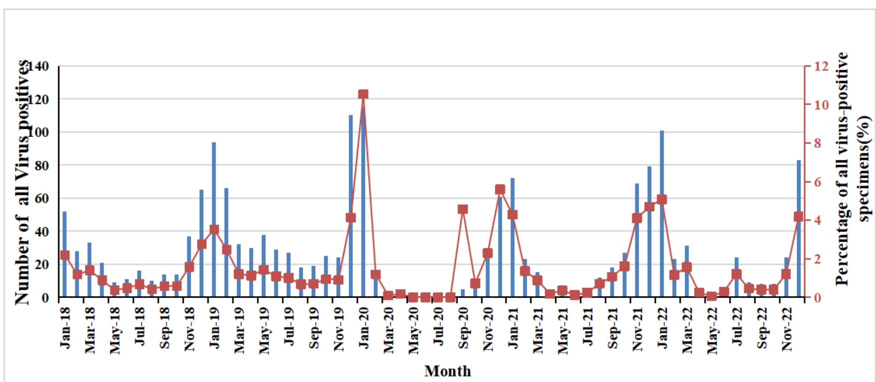
virus infection, B:The number of positive and positive rates of RSV infection,C:The number of positive and positive rates of ADV infection, D:The number of positive and positive rates of Flu A infection, E:The number of positive and positive rates of Flu B infection, F:The number of positive and positive rates of PIV-1 infection, G:The number of positive and positive rates of PIV-2 infection, H:The number of positive and positive rates of all respiratory virus infection.

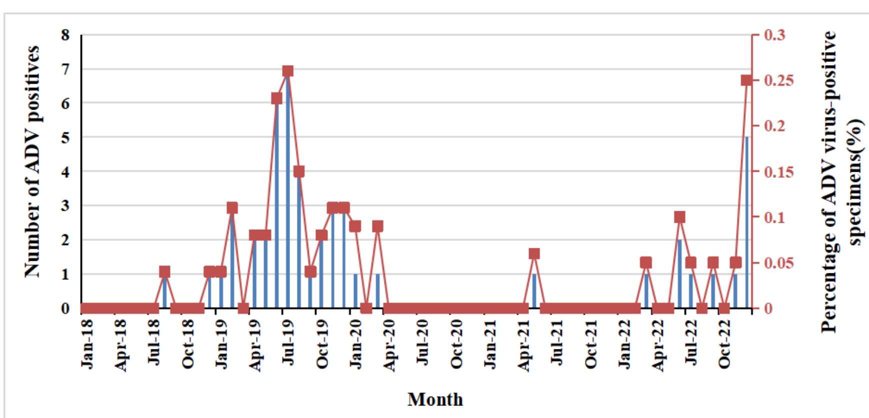
Figure 2. Seasonal distribution trends of respiratory virus infection in hospitalized children with acute respiratory infection, 2018-2019 and 2020-2022

Figure 1

A B







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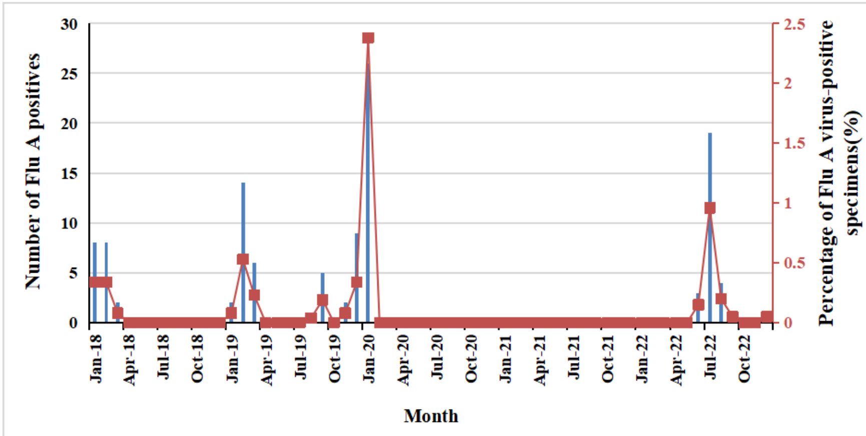
Number of Flu A positives
0
5
10
15
20
25
30
Percentage of Flu A virus-positive specimens(%)
0
0.5
1
1.5
2
2.5
Jan-18
Apr-18
Jul-18
Oct-18
Jan-19
Apr-19
Jul-19
Oct-19
Jan-20
Apr-20
Jul-20
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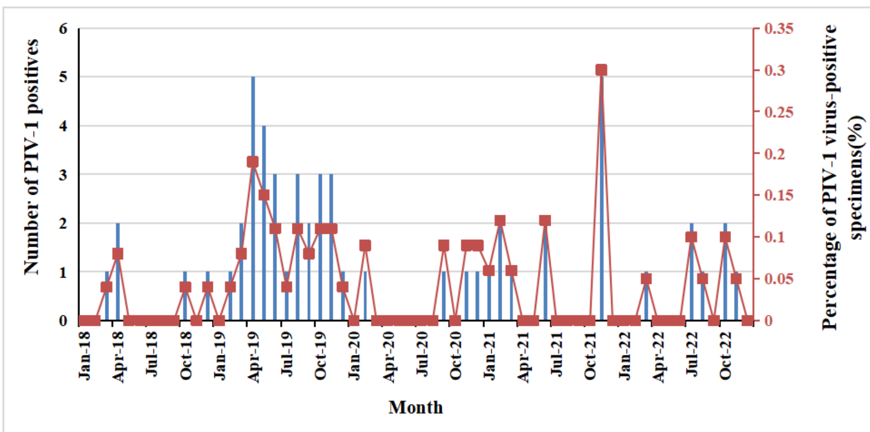
Oct-22
Month

E

E F


Number of PIV-1 positives
0
1
2
3
4
5
6
Percentage of PIV-1 virus-positive specimens(%)
0
0.05
0.1
0.15
0.2
0.25
0.3
0.35
Jan-18
Apr-18
Jul-18
Oct-18
Jan-19
Apr-19
Jul-19
Oct-19
Jan-20
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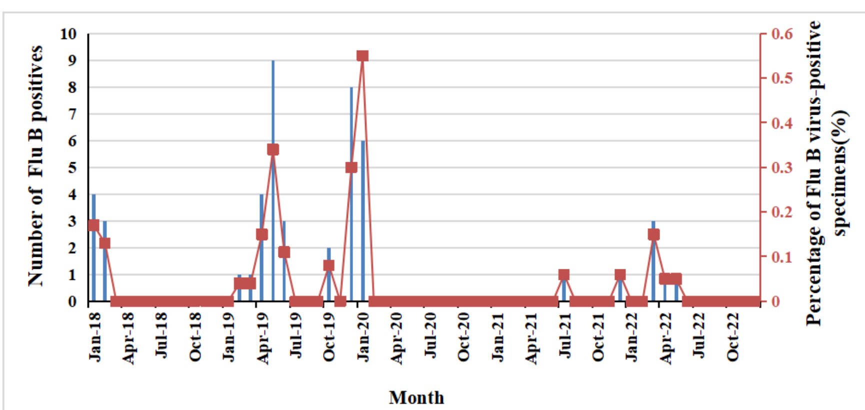
Jul-22
Oct-22
Month


Number of Flu B positives
0
1
2
3
4
5
6
7
8
9
10
Percentage of Flu B virus-positive specimens(%)
0
0.1
0.2
0.3
0.4
0.5
0.6
Jan-18
Apr-18
Jul-18
Oct-18
Jan-19
Apr-19
Jul-19
Oct-19
Jan-20
Apr-20
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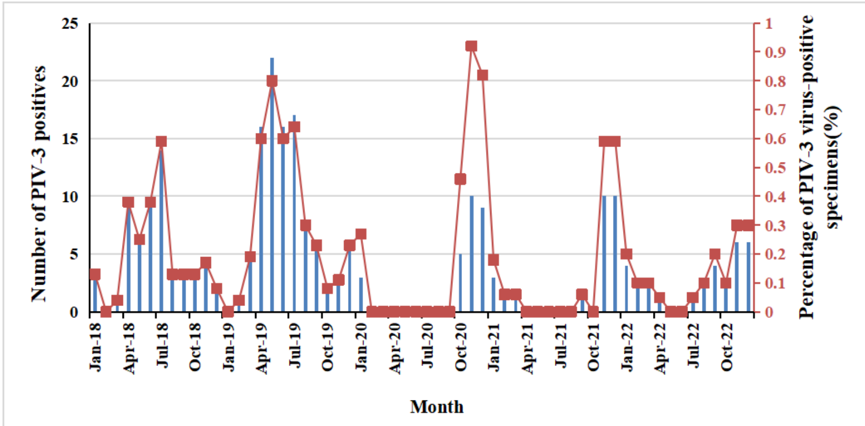
Oct-22
Month

G H



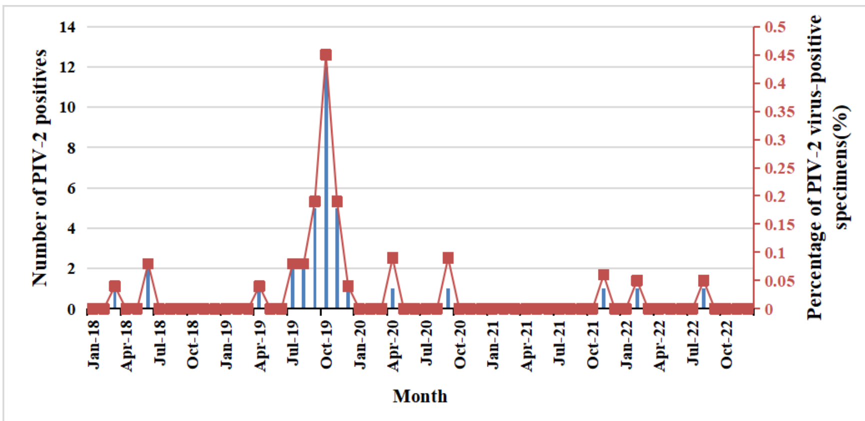
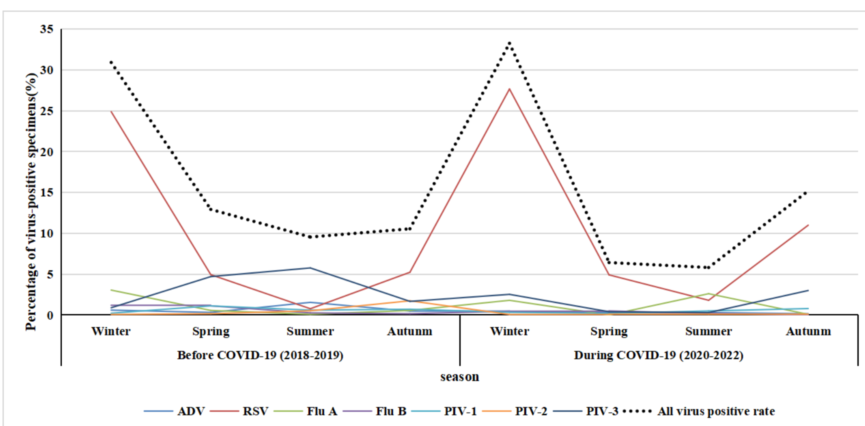


Figure 2



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