

Epidemiology of Respiratory Viruses Detected by Multiplex Real-time PCR in Hospitalized Patients with Lower Respiratory Tract Infections in Vietnam from 2020 to 2024

Huong Thien Pham^{1,2,3*}, Van Hung Pham^{1,3}, Nhu Hong Thi Tran^{1,3}, Duy Khanh Tran^{1,3}, Thao Hanh Thi Nguyen^{1,3}, Quang Duy Ha^{1,3}, Anh Hong Pham^{1,2,4}
¹Vietnam Research and Development Institute of Clinical Microbiology, Ho Chi Minh city 70000, Vietnam; ²Pham Ngoc Thach University of Medicine, Ho Chi Minh city 70000, Vietnam; ³Nam Khoa Biotek Co

Background

Respiratory tract infections (RTIs) remain a leading cause of global hospitalization and mortality. The COVID-19 pandemic significantly disrupted traditional viral circulation and seasonal patterns. Large-scale surveillance using advanced techniques, such as multiplex real-time PCR (MLP-rPCR), is essential for preventive strategies and antimicrobial stewardship.

Aims of the study

This study aimed to assess viral detection trends in hospitalized RTI cases in Vietnam from 2020 to 2024 and highlight the necessity of comprehensive molecular screening.

Materials and methods

We conducted a five-year retrospective analysis of 15,936 respiratory clinical samples from hospitalized patients. The workflow of viral detection using an in-house MLP-rPCR assay targeting 17 respiratory viruses is shown below.

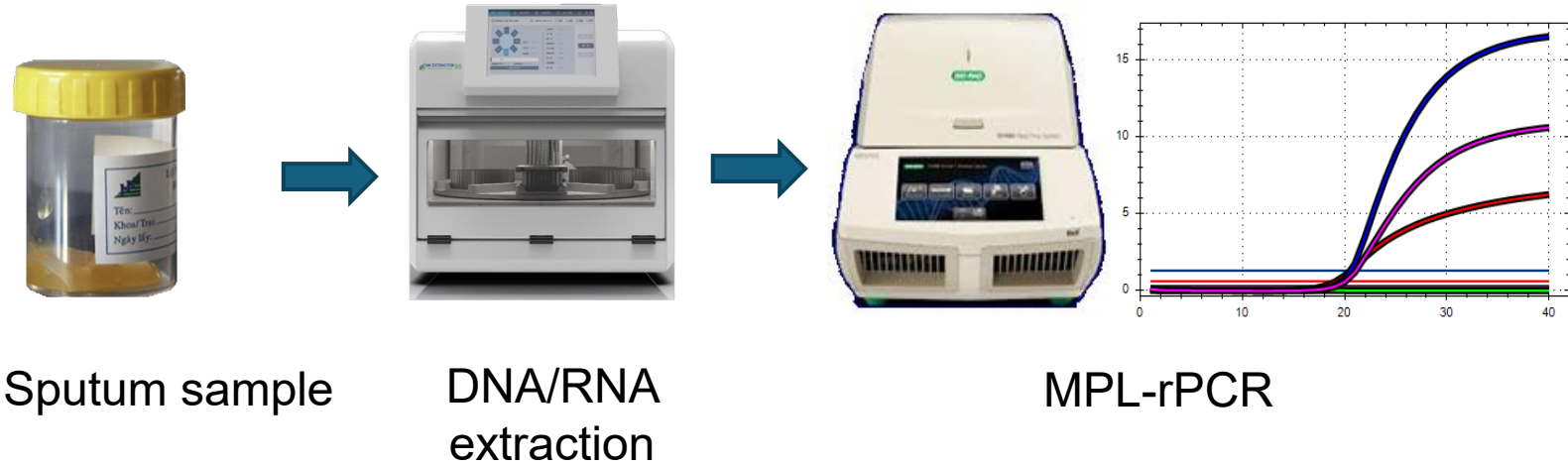


Figure 1: Flow chart of the real-time PCR for detecting viruses causing respiratory infection in sputum

Table 1. Respiratory viruses detected by MPL-rPCR assay

Influenza A (InfA)	Parainfluenza virus 1 (Para1)	Respiratory syncytial virus (RSV)
Influenza B (InfB)	Parainfluenza virus 2 (Para2)	Human adenovirus (ADV)
Influenza C (InfC)	Parainfluenza virus 3 (Para3)	Measles virus (MeaV)
Human metapneumovirus (hMPV)	Human coronavirus HKU1 (hCoV)	SARS-CoV (SARS)
Rhinovirus (Rhino)	SARS-CoV-2 (CoV2)	MERS-CoV (MERS)
Bocavirus (Boca)	Varicella-zoster virus (VZV)	Rubella virus (RUBV)

Results

Viral pathogens were identified in 31.9% of specimens, higher in children than in adults

Table 2. Number and percentage of respiratory viruses detected in hospitalized RTI cases among adults (≥16 years) and children (<16 years)

Respiratory Virus	Children N (%)	Adults N (%)	χ ²	p
InfA	341 (10.22)	511 (29.28)	298.3	<0.001
InfB	52 (1.56)	86 (4.93)	49.23	<0.001
InfC	19 (0.57)	11 (0.63)	0.07	>0.05
Para1	65 (1.95)	10 (0.57)	14.9	<0.001
Para2	38 (1.14)	9 (0.52)	4.86	>0.05
Para3	561 (16.82)	148 (8.84)	66.29	<0.001
RSV	733 (21.97)	106 (6.07)	210.05	<0.001
ADV	594 (17.81)	110 (6.30)	126.99	<0.001
MeaVR	278 (8.33)	5 (0.29)	141.05	<0.001
Rhino	775 (23.23)	273 (15.64)	40.28	<0.001
hMPV	153 (4.59)	77 (4.41)	0.08	>0.05
Boca	542 (16.25)	31 (1.78)	239.77	<0.001
hCoV	9 (0.27)	3 (0.17)	0.143*	>0.05
CoV2	94 (2.82)	447 (25.62)	625.93	<0.001
VZV	3 (0.09)	7 (0.40)	4.176*	>0.05
Respiratory virus [+]	3336 (52.98)	1745 (18.10)	2132.9	<0.001

Statistical significance was defined as $p < 0.05$

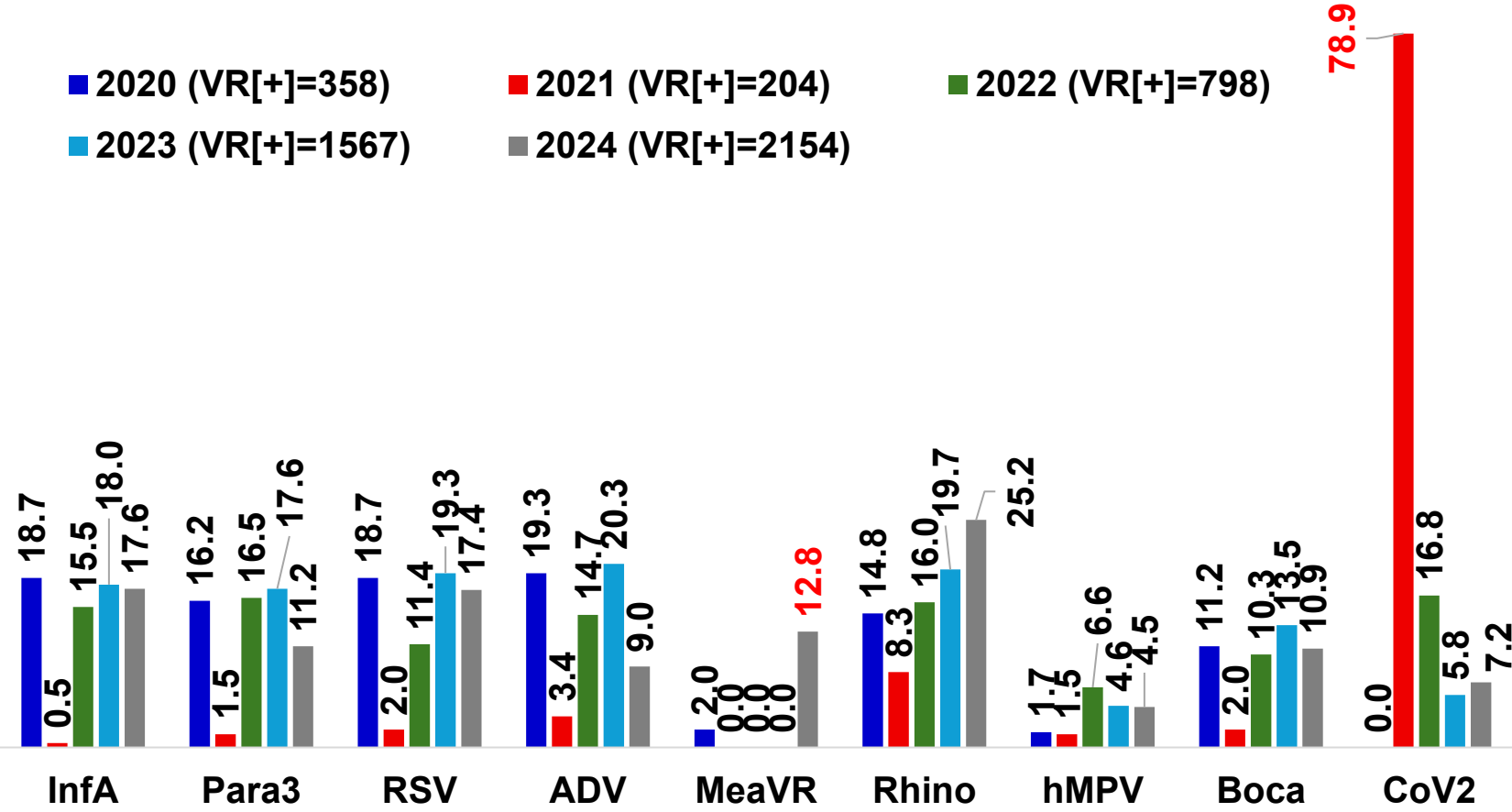


Figure 3. Detection rates (%) of nine common respiratory viruses from 2020 to 2024. During the 2021 pandemic peak, CoV-2 accounted for nearly 79% of detections, suppressing other respiratory viruses. By 2024, a significant MeaVR re-emergence was observed in children (17.7%).

Conclusions

This study demonstrates a significant shift in the respiratory viral circulation in Vietnam following the COVID-19 pandemic. MLP-rPCR is a practical and reliable tool for resource-limited settings. Routine molecular testing can enhance diagnostic accuracy, guide clinical management, and strengthen public health surveillance.

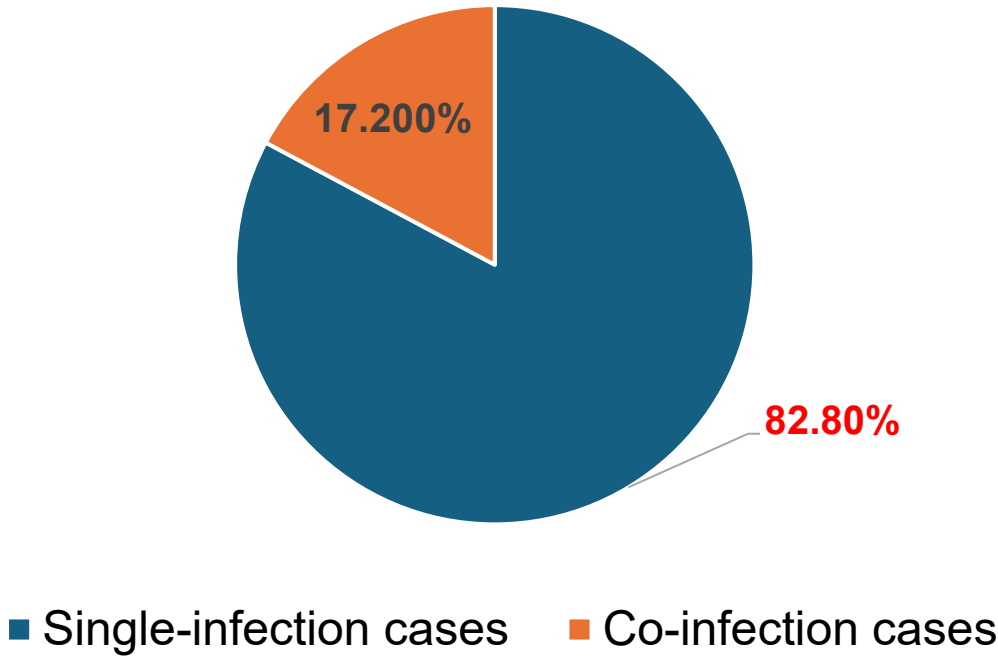


Figure 2. Cases and percentages of single viral infections among respiratory viruses detected.

Table 3. Monthly detection rates (%) of respiratory viruses in 2023 and 2024 based on 9319 samples from the South of Vietnam. Circulation varied throughout the year, with InfA and RSV prevalent from February to November and May to November, respectively. In contrast, Para3, ADV, hMPV, and Boca peaked during the late-year and early-year months (October to April). Rhino circulated broadly year-round, while CoV2 was concentrated between April and June.

	InfA	Para3	RSV	ADV	MeaVR	Rhino	hMPV	Boca	CoV2	VR[+]
Results from 9319 samples in the South of Vietnam										
Jan	4.21	25.89	2.59	25.89	0.00	36.25	5.50	20.39	5.83	44.02
Feb	15.54	23.90	5.18	19.12	0.00	37.85	5.58	13.94	2.39	38.20
Mar	15.14	23.11	6.37	15.94	0.00	36.65	2.79	12.75	2.39	34.86
Apr	14.97	15.51	13.90	12.83	1.07	19.25	4.81	18.18	11.76	30.41
May	9.71	8.74	25.73	10.19	2.43	19.42	0.00	9.71	29.13	29.30
Jun	16.30	8.70	30.98	3.80	4.35	15.22	3.26	11.96	19.57	28.66
Jul	22.62	5.43	35.75	5.43	4.98	24.89	1.36	9.95	6.79	30.27
Aug	33.18	2.84	29.86	9.48	9.95	9.95	0.95	9.48	5.21	30.49
Sep	18.14	6.51	35.35	5.58	10.70	24.65	2.33	9.77	0.47	33.75
Oct	25.25	4.70	32.67	11.88	7.92	16.09	4.95	6.44	1.24	42.26
Nov	24.14	11.36	16.84	14.00	13.59	14.81	7.10	9.94	2.23	44.45
Dec	8.89	22.59	8.70	17.96	19.26	22.41	7.96	15.19	4.26	46.71

References

- Ly, V.K.; Pham, V.H.; Van Ly, X.; Pham, P.M. Bacterial and viral co-infections in community-acquired pneumonia in adults: a prospective study of multiple hospital centers. *MedPharmRes* **2024**, *8*, 153–161. <https://doi.org/10.32895/UMP.MPR.8.3.17>.
- Tang, Z.; Fan, H.; Tian, Y.; Lv, Q. Epidemiological characteristics of six common respiratory pathogen infections in children. *Microbiol. Spectr.* **2025**, *13*, e0007925. <https://doi.org/10.1128/spectrum.00079-25>.
- Burk, M.; El-Kersh, K.; Saad, M.; Wiemken, T.; Ramirez, J.; Cavallazzi, R. Viral infection in community-acquired pneumonia: a systematic review and meta-analysis. *Eur. Respir. Rev.* **2016**, *25*, 178–188.