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Molecular detection of Human Metapneumovirus among geriatric patients presenting with acute respiratory tract infection in Ibadan, Nigeria

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Abstract

Acute respiratory tract infections (ARTIs) are among the leading causes of morbidity and mortality worldwide, and viruses are primarily responsible for these infections. The Human Metapneumovirus (hMPV) is a respiratory pathogen that was first identified in 2001 in the Netherlands, and has since been recognized as one of the major viral causes of respiratory infections. The clinical manifestations of hMPV infection are typically asymptomatic or mild in healthy adults, however, for high-risk populations, such as children less than 5 years, older people, immunocompromised individuals, and patients with underlying chronic conditions, hMPV infection can lead to severe clinical manifestations, such as pneumonia.

Despite its substantial health burden, hMPV is not routinely tested for and monitored in many countries, there are also no approved vaccines or antiviral treatments, and management of the infection remains supportive. The recurrent hMPV related outbreaks in recent years and its potential to cause pandemics, particularly in the post-COVID-19 era, highlights the urgent need for improved surveillance and targeted interventions.

Oropharyngeal and nasal swab samples were collected from five hundred and twenty four (524) geriatric patients comprising of outpatient individuals presenting with symptoms of acute respiratory tract infection (ARTI) and those attending the clinic for routine health checks recruited as asymptomatic controls from the period of April 2024 to May 2025. The samples were tested for the presence of Human metapneumovirus using a semi-nested Polymerase chain reaction targeting the Nucleoprotein (N) gene of the virus.

Thirty six (6.9%) of the 524 samples were positive for Human Metapneumovirus infections with a significantly ($p = 0.004$) higher rate among symptomatic participants (5.2%). The difference in the rate of hMPV positivity between age groups and gender was not statistically significant ($p = 0.707$; $p = 0.344$ respectively). The three symptoms most commonly reported among hMPV positive cases in our study were cough (30.6%), fever (25%), and headache (25%); however the comparison of clinical symptoms across all the participants showed that chills and sore throat were the only symptoms statistically significant with hMPV positivity ($p = 0.006$; $p = 0.023$ respectively). HMPV positivity peaked during the month of July with the highest number of infections detected in symptomatic and asymptomatic individuals within that month.

This study highlights the impact of hMPV among the geriatric population in Ibadan, Nigeria, indicating a necessity for more accurate and dependable estimates of the incidence of hMPV-associated disease in the older adult population.

Introduction

Acute respiratory tract infections (ARTIs) are among the leading causes of morbidity and mortality worldwide and viruses are responsible for a significant proportion of these infections. Among individuals

with ARTIs, the elderly, due to atypical and delayed presentation, co-morbidities, aging immune system, suffer disproportional morbidity and mortality rates, from viral respiratory infections. Before the COVID-19 pandemic, 90% of influenza-related deaths were reported to occur in the elderly (Talbot *et al*, 2011). Severe acute respiratory syndrome-Coronavirus-2 (SARS-CoV-2) contributed to this high burden as more severe COVID-19 disease was observed particularly among older persons, Number of deaths in a U.K study due to COVID-19 increased with 12.8%, 29.2% and 50.4% of deaths seen in 60–69, 70–79 and 80 + year olds, respectively(Docherty *et al*, 2020). Other respiratory viruses including the Human Metapneumovirus (hMPV) and Human Respiratory syncytial virus (HRSV) are also being recognized as etiological agents causing substantial morbidity and mortality in the elderly(Thompson *et al*, 2003).

HMPV is a respiratory pathogen that was first identified and reported in 2001 in the Netherlands (Van Hoogen *et al*, 2001) and has since been recognized as one of the major viral causes of respiratory infections. Although HMPV often causes moderate to self-limiting respiratory tract infections, it can also cause bronchiolitis and severe pneumonia, particularly in older adults, small children, and immune-compromised patients (Kulkarni *et al*, 2025). HMPV is an enveloped, single-stranded, non-segmented RNA virus with a genomic size ranging between 13,281 and 13,387 nucleotides in length. It is a member of the Metapneumovirus genus, of the *Pneumoviridae* family. Its genome consists of single-stranded, negative-sense, non-segmented ribonucleic acid (RNA) that encodes nine proteins across eight genes: nucleoprotein (N), phosphoprotein (P), matrix protein (M), fusion protein (F), matrix-2 proteins (M2-1 and M2-2), small hydrophobic (SH) protein, glycoprotein (G), and the large polymerase protein (L) (Panda *et al*, 2014).

Since its discovery, HMPV has been reported on every continent, confirming its widespread occurrence and importance to public health. Despite the global prevalence and spread of HMPV, there are currently no approved vaccines or antiviral treatments against the virus. The resurgence of HMPV in China in recent years (2024–2025) has brought attention to the virus's potential effects on public health and further raised fears on the possibility of the virus to cause a pandemic. While HMPV was once considered a less prominent cause of respiratory infections, recent outbreaks, particularly in non-endemic regions, have highlighted its potential to cause widespread illness. In light of the aging global population and the increasing burden of chronic comorbidities, there is a pressing need for current, targeted evidence on the incidence, burden, clinical presentation, and healthcare impact of hMPV infection among vulnerable groups.

In this study, the prevalence of hMPV infection among older persons (≥ 60 years) was determined using reverse transcription and semi- nested Polymerase chain reaction (PCR) for detection of hMPV Nucleoprotein gene from oropharyngeal and nasal swab specimens.

Materials and methods

Participant enrollment and sample collection

A total of 524 geriatric outpatients, aged 60 years and older, who provided their consent, were enrolled in the study. This group included 266 individuals who had symptoms of acute respiratory tract infections for no longer than seven days (WHO, 2012) and other elderly patients with no symptoms of ARTI(258 individuals) visiting the clinic for routine health checks.. Recruitment took place at three hospitals: the geriatric clinic of the University College hospital, Ibadan; Adeoyo hospital, Yemetu and Primary health center, Idi-Ogungun all located in Ibadan North, in South-west Nigeria, from April 2024 to May 2025. A structured questionnaire was administered to each participant to collect demographic and clinical information. The clinical symptoms included fever, cough, chills, nasal congestion, sore throat, loss of taste or smell, headache, fatigue, palpitations, conjunctivitis, and chest pain. A combination of oropharyngeal and nasal swab samples was collected from each patient, both swabs were placed together in a cryovial containing 500uL of DNA/RNA shield, transported on ice to the laboratory and stored at − 80°C until analysed.

Ethical approval for the study was obtained from the University of Ibadan/University college hospital (UI/UCH) ethics review board with approval number UI/EC/24/0167. In addition, approval was also received from the Oyo state ethics review board (approval number: NHREC/OYOSHREC/10/11/22).

RNA extraction and RT-PCR detection of HMPV N gene

RNA was extracted from 140 µl of the swab sample using the Qiagen total RNA isolation kit according to the manufacturer's instructions and samples were stored at -80^0 C prior to amplification reactions. Nested Polymerase chain reaction (PCR) was used to amplify the nucleoprotein (N) gene of hMPV (N gene has been reported by Cote *et al*, 2014 as one of the most suitable genes for hMPV detection with high specificity and sensitivity), using Superscript III one step RT-PCR platinum Taq HiFi. The thermal cycling conditions of the first-round PCR was 50^{0}C for 30 min (the reverse transcription step), 94^{0}C for 15 min, 35 cycles of 94^{0}C for 30 seconds, 57^{0}C for 10 seconds, 72^{0}C for 40 seconds, and a final extension at 72^{0}C for 5 min. The product of the first round PCR was used for the second round at the following thermal cycling conditions: 95^{0}C for 3 minutes, 35 cycles of 95^{0}C for 30 seconds, 56^{0}C for 30 seconds, 72^{0}C for 40 minutes, and final extension at 72^{0}C for 5 min, using the 5X Firepol master mix (Solis Biodyne, Europe). Non-template controls as well as the positive control (HMPV PZ065611) was included in all of the assays. The sequences of the specific primers used are listed in Table 1. The PCR products were visualized on a 1.5% ethidium bromide-stained agarose gel, and size was discriminated using a 100 bp Plus DNA ladder (Solis biodyne, Europe).

Table 1
Sequences of primers used in amplification of hMPV

Virus	Virus target	Primer sequence	Amplicon size(bp)
hMPV	N gene	Forward: AAGCATGCTATATTAAAAGAGTCTCA	440
		Reverse: ATTATGGGTGTGTCTGGTGCTGA	365
		Internal reverse: ATTGTTTTTCTTGCTTCTTTGTCTAT	

Statistical analysis

Statistical analyses were performed using Excel and SPSS and significant levels were calculated by Chi-square method. Data were displayed as numbers or percentages (%). The "*P*" value > 0.05 was employed to evaluate statistical significance.

Results

Thirty six of the 524 (6.9%) samples analyzed were positive for hMPV Nucleoprotein (N) gene of the virus. Figures 1 and 2 shows the amplified hMPV specific N gene in participant's samples (see supplementary file).

The rate of hMPV infection was significantly higher (p = 0.004) among the symptomatic (10.2%) than the asymptomatic (3.5%) group (Fig. 3).

Table 2
HMPV infection by demographic characteristics of the study population

Variable	Values			P-value
		No. tested	No. Positive (%)	
Age_group	60–69	252	18(7.1)	0.707
	70–79	205	15(7.3)	
	≥ 80	67	3(4.5%)	
Total		524	36(6.9%)	
Sex	Female	398	25(6.3%)	0.344
	Male	126	11(8.7%)	
Total		524	36(6.9%)	
Study Site	Ade-Oyo	77	6(7.8%)	0.91
	PHC	67	5(7.5%)	
	UCH	380	25(6.6%)	
Total		524	36(6.9%)	
Status	Symptomatic	266	27(10.2%)	0.004*
	Asymptomatic	258	9(3.5%)	
Total		524	36(6.9%)	

Although the rate of hMPV was higher among the 70–79 age group (7.3%), there was no significant difference (p = 0.707) by age (Table 2). Similarly, the infection was also higher among male participants (Fig. 4), however, this difference was not significant (p = 0.344).

The three symptoms most commonly reported among hMPV positive participants in this study were cough (30.6%), fever (25%), and headache (25%). Further analysis of respiratory symptoms showed that chills and sore throat were the only symptoms statistically associated (p = 0.006 and 0.023 respectively) with hMPV positivity (Table 3).

Table 3
Frequency of respiratory symptoms among hMPV positive geriatric patients

Symptom	Overall (N = 524)	HMPV positive	HMPV negatives	P value
Cough	121 (23.09%)	11 (30.6%)	110 (22.5%)	0.271
Fever	106 (20.23%)	9 (25.0%)	97 (19.07%)	0.46
Headache	118 (22.52%)	9 (25.0%)	109 (22.3%)	0.906
Chills	49 (9.35%)	8 (22.2%)	41 (8.4%)	0.006**
Sore Throat	57 (10.88%)	8 (22.2%)	49 (10.04%)	0.023**
Fatigue	84 (16.03%)	6 (16.7%)	78 (16.0%)	0.757
Catarrh/Runny Nose	58 (11.07%)	5 (13.9%)	53 (10.9%)	0.576
Conjunctivitis	50 (9.54%)	4 (11.1%)	46 (9.4%)	0.326
Palpitations	47 (8.97%)	4 (11.1%)	43 (8.8%)	0.719
Loss of Taste/Smell	39 (7.44%)	3 (8.3%	36 (7.4%)	0.833
Chest Pain	33 (6.30%)	3 (8.3%)	30 (6.2%)	0.795
Shortness of Breath	28 (5.34%)	1 (2.7%)	27 (5.5%)	0.519
Total	524	36	488	
** Statistically significant				

Analysis by month of sample collection (Table 4) showed that the highest prevalence of hMPV infections was found in the month of July 2024 (30.6%) followed by May 2025 (21%), however, there was no clear pattern in the detection of the virus by month. No hMPV was detected in the months of September and October in 2024 and January, February and April 2025. It is worthy of note that the peak of hMPV detection in 2024 was same for both symptomatic and asymptomatic individuals(July and November-December 2024).In 2025, there was no hMPV infection in the controls, while infection peaked in March and May for the symptomatic individuals (Fig. 5).

Table 4
ASSOCIATION BETWEEN HMPV POSITIVITY AND MONTHS OF SAMPLE COLLECTION

Month	HMPV+	Total	% Positive	Chi-square p-value
Apr-24	2	57	3.51%	*P < 0.001*
May-24	1	92	1.09%	
Jun-24	1	26	3.85%	
Jul-24	11	36	30.56%	
Aug-24	4	29	13.79%	
Sep-24	0	12	0.00%	
Oct-24	0	24	0.00%	
Nov-24	7	57	12.28%	
Dec-24	5	40	12.50%	
Jan-25	0	46	0.00%	
Feb-25	0	39	0.00%	
Mar-25	2	23	8.70%	
Apr-25	0	29	0.00%	
May-25	3	14	21.43%	
Total	36	524	100%	

Discussion

Respiratory tract infections resulting from hMPV is a significant health issue for infants, young children, the elderly and immune-compromised individuals. In this cross sectional study an overall hMPV infections prevalence of 6.9% was obtained among elderly persons (≥ 60 years).

This rate is higher than some earlier reports of hMPV positivity in ARI cases from studies in India, Banerjee *et al*,2011(3.7%); Kenya, Owor *et al*, 2016(4.8%); Senegal, Dia *et al*, 2014(3.0%); Malaysia, Chow

et al, 2016 (2%);China, Zhao *et al*, 2022(5.3%); Mu *et al*, 2024(3.06%); Bangladesh, Ul Islam *et al*,2024(5.08%); and Saudi Arabia Almuqrin et al, 2025(4.3%); but lower than the prevalence of 12.07% which was reported in a study among rural and peri-urban children in South India by Naranayan *et al* in 2016.

The difference in prevalence of hMPV infections in these studies and ours may be due to differences in the population studied, climate, patient's age and geographical region. Of particular note is that several of these studies were either in children or general population while our study focused on elderly person's aged ≥ 60 years, who were mostly outpatients. Globally, the hMPV prevalence in hospital inpatient and community studies, in children and the elderly, varies from 1.7 to 17%, with a higher prevalence in outpatients compared to inpatients, and in children < 5 years of age compared to older age groups (Banerjee et al, 2011; Williams *et al*, 2010). In addition, the presence of atypical symptoms, higher exposure to environments where respiratory infections are prevalent, such as healthcare settings and lack of routine testing for non-Influenza viruses among the elderly may also contribute to varying rates of infection.

The 3.5% prevalence of hMPV found among asymptomatic elderly participants in Ibadan, Nigeria aligns with rates reported in other places (Sobanjo-ter Meulen *et al*, 2025) among similar population group with prevalence of hMPV in asymptomatic older adults ranging from 0.0% to 3.4% across three studies carried out in the USA(Walsh, 2008) and Netherlands(van Beek, 2017; Kaajik, 2022). This rate is crucial because it serves as a key indicator for assessing the actual prevalence of respiratory viruses (Wang TE et al. 2010), given that asymptomatic carriers play a significant role in the transmission of the infection. Although there is evidence that viral shedding correlates with symptom severity and that the contagiousness of asymptomatic individuals is less than for symptomatic persons (WHO, 2006), the absence of symptoms might bring asymptomatic infected individuals into greater contact with susceptible persons in and outside of the home.

Although there was no significant association between gender and hMPV positivity, the higher proportion of infected male participants observed in this study is similar to the findings in previous studies by Yi *et al*, 2019 (China) and Almuqrin et al, 2025 (Saudi-Arabia) who reported a pre-dominance of hMPV infections among males. The male predominance could be due to various socio-cultural factors, such as occupational exposure in their early ages which makes them more susceptible to infectious agents later in their geriatric ages due to a compromised respiratory system; and also the reluctance of male elderly individuals to seek medical care until symptoms have set in.

Based on our findings in this study, chills and sore throat showed a significant correlation with hMPV positivity in this group; however, we noted a high prevalence of cough and fever among hMPV positive participants, with cough being the most frequently reported symptom. This aligns with earlier findings in patients infected with hMPV, as Hamelin et al. (2005) reported cough in 100% of hMPV-infected patients; Walsh et al. (2008) noted cough in 94% of those who tested positive; Loubet et al. (2020) found fever present in 88% of hMPV positive individuals, and Philipot et al. (2024) found cough in 85% of those who

were hMPV positive. These symptoms although not specific for hMPV since similar observations have been reported with other respiratory viruses which cause upper respiratory tract infections (Jennings *et al*, 2018) highlight the need to test for other respiratory viruses apart from Influenza in ARI cases among the elderly in order to avoid delayed or missed diagnosis.

In our study, HMPV was detected from April to August and then in November to December in the year 2024 with a noticeable peak in July (which falls within Nigeria's rainy season) and November to December in both symptomatic and asymptomatic individuals. The notable rise in infections observed in July corresponds with previous findings from tropical Pune in central India, where hMPV detections were highest during July and August in the year of study (Rao *et al.*, 2004), as well as reported peaks of hMPV cases occurring in June and July in the southern hemisphere (Panda *et al*., 2014). In Kenya, peak HMPV prevalence has been recorded in June-July in the west and November-December in refugee camps in the northeast and northwest of the country (Ahmed et al, 2012; Feikin *et al*, 2012).

Earlier research has indicated that hMPV in sub-tropical regions like India, was detected from September through April, with a noticeable peak of detections in March of the year of study (Banerjee et al, 2011). In China, HMPV infections were reported to peak from November to February (Mu *et al*, 2024); and in a 10-year retrospective study by Ye et al, 2023, over half of the hMPV infections were detected in spring (March to May), an occurrence which coincides with the virus detection among symptomatic individuals in our study during the year 2025(Fig. 5).

This varying pattern of hMPV circulation may be due to climatic and demographic differences as climate plays an important role in determining the seasonality of HMPV in many tropical countries. Previous studies have described a direct correlation between the number of rainy days and the increase in the number of HMPV infections (Chow *et al*, 2016) which is in line with the findings from our study. In addition to the number of rainy days, the effects of other important factors in a tropical climate such as the relative humidity, temperature and amount of rainfall on the prevalence of HMPV need to be investigated in order to provide a better understanding of its seasonality.

The samples used in this study were collected over a period of 13 months, and this is a limitation of the study because no clear peak patterns were observed. To accurately determine the seasonality of hMPV and other respiratory viruses requires collection of samples over multiple years to establish a trend or pattern. Currently, there is only a limited amount of information regarding hMPV in the general population of Nigeria, and this article to the best of our knowledge is the first to present data on the prevalence of hMPV infection in this cohort. The findings on hMPV from this research reveals the need for continuous surveillance among the geriatric population nationwide to evaluate the disease burden and gain a clearer understanding of its epidemiology and clinical relevance.

Conclusion

The findings from this study, offers significant insights into hMPV prevalence in the elderly. To date, the majority of research has concentrated on the molecular epidemiology of HMPV in hospitalized children,

resulting in a gap in understanding the prevalence and genotypic distribution among the broader population across all age groups.

Along with previous studies, the findings from this research indicate that the impact of hMPV on older adults is significant and clinically important. Therefore, it is crucial to establish more accurate and dependable global estimates of the burden due to hMPV in elderly persons. Continued surveillance of hMPV-associated ARIs is necessary to understand the epidemiology of the virus in the aftermath of the COVID-19 pandemic. Such evidence will help guide future vaccination strategies to ensure optimal protection from hMPV-associated ARIs in elderly persons.

Declarations

Ethical approval and consent to participate

Ethical approval for this study was obtained from the University of Ibadan/University college hospital (UI/UCH) ethics review board with approval number UI/EC/24/0167. In addition, approval was received from the Oyo state ethics review board (approval number: NHREC/OYOSHREC/10/11/22).Informed consent was obtained from all participants prior to their inclusion in the study.

Consent for Publication

Not Applicable

Data availability

The data supporting the conclusions of this article are available within the article and/or its Supplementary materials. Further data will be provided by the corresponding author upon request.

Declaration of Helsinki

This study adhered to the Declaration of Helsinki as consent was obtained from all individuals before obtaining their samples.

Competing Interests

The authors of this paper declare that they have no competing interests.

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Author's Contribution

ATJ – Conceptualization, investigation, data curation, methodology, project administration, validation, visualization, writing—original draft preparation, writing—review and editing, resources and fund

acquisition.

AOF– Resources, fund acquisition, Supervision, writing—review and editing.

GNO – Resources, fund acquisition, Supervision, writing—review and editing.

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Figures

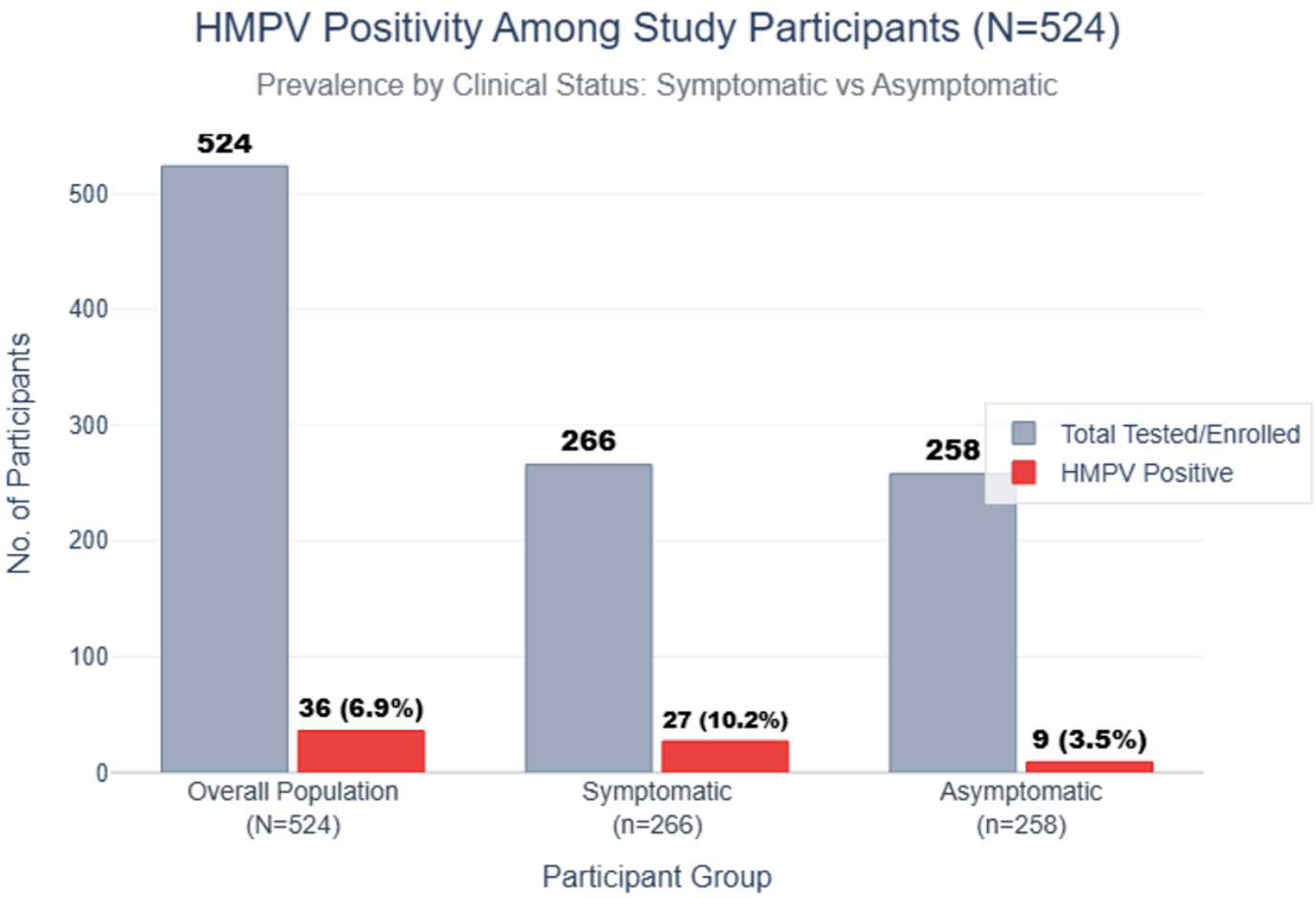


Figure 1

3: HMPV Positivity among the study participants.

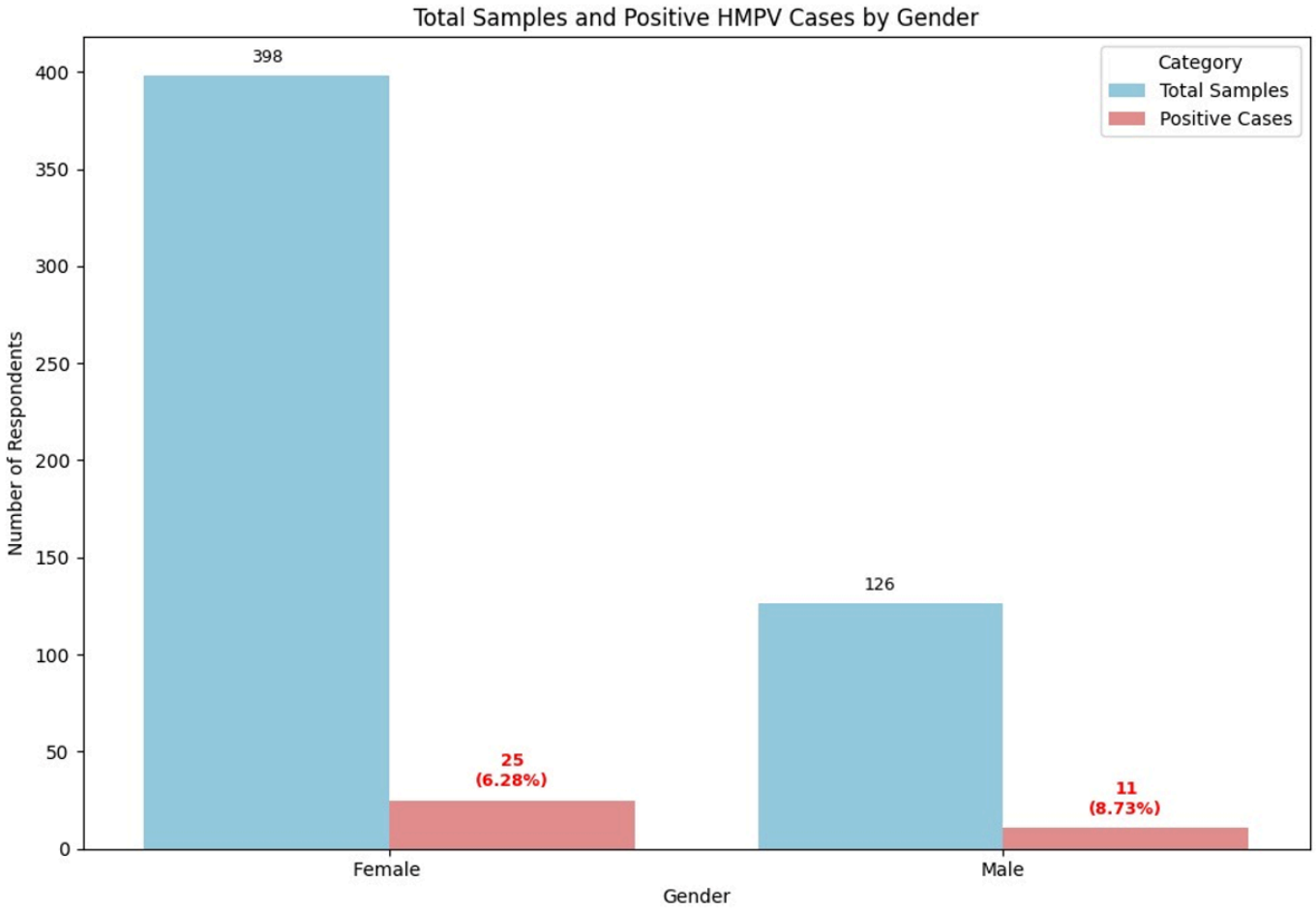


Figure 2

4: HMPV positivity among the participants based on gender.

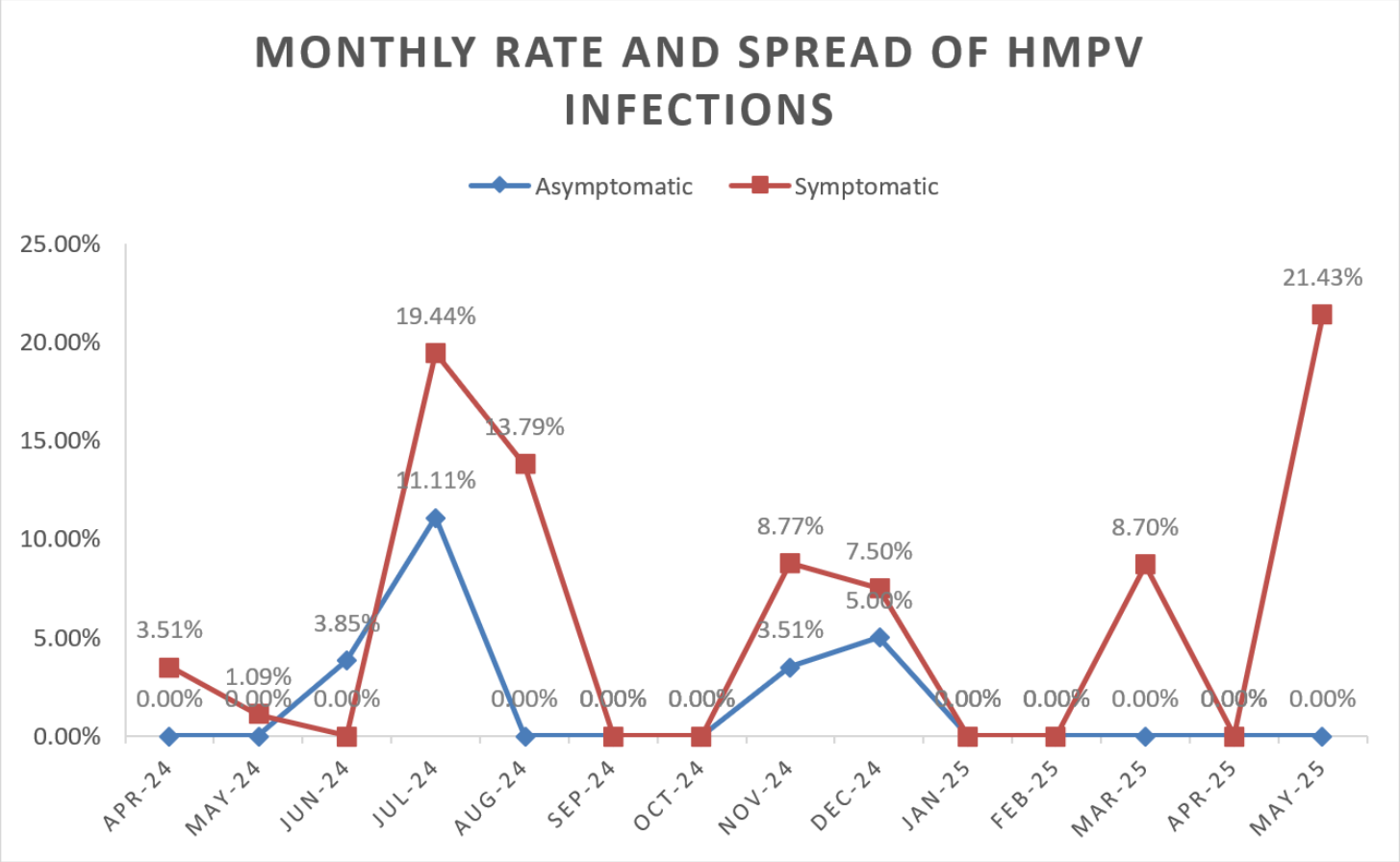


Figure 3

5: Monthly distribution of HMPV infection among symptomatic and asymptomatic participants.

Supplementary Files

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- SupplementaryDataBMC.docx