

Correlation Between Inflammatory Markers and Pathogenic Bacteria in Children's Winter Respiratory Infections in Xinjiang

Lixia Wang^{1,*}, Wei Wang^{2,*}, Jian Wang¹, Ling Zhu¹, Jianjiang Luo¹

¹Department of Respiratory and Critical Care Medical Department Infectious Diseases Ward, The Traditional Chinese Medicine Hospital Affiliated to Xinjiang Medical University, Urumqi, Xinjiang, People's Republic of China; ²Department of Respiratory and Critical Care Medicine, Affiliated Traditional Chinese Medicine Hospital of Xinjiang Medical University, Urumqi, Xinjiang, People's Republic of China

*These authors contributed equally to this work

Correspondence: Jianjiang Luo, Department of Respiratory and Critical Care Medical Department infectious diseases Ward, The Traditional Chinese Medicine Hospital Affiliated to Xinjiang Medical University, No. 116 Huanghe Road, Shayibake District, Urumqi, Xinjiang, People's Republic of China, Tel +86013999946655, Email ljj.781201@163.com

Background: This study examines the distribution characteristics of pathogenic bacteria in respiratory infections and their relationship with inflammatory markers to guide clinical drug use.

Methods: We selected 120 patients with lower respiratory tract infection in the electronic medical record system of Xinjiang Provincial People's Hospital from March 2019 to March 2023 for a case-control study. Using Indirect Immunofluorescence Antibody test (IFA), blood routine, C-reactive Protein (CRP), and High-sensitivity C-reactive Protein (hsCRP), we detected nine respiratory pathogens (Respiratory syncytial virus; Influenza A virus; Influenza B virus; Parainfluenza virus; Adenovirus; Mycoplasma pneumoniae; Chlamydia pneumoniae; Legionella pneumophila type 1; Rickettsia Q) in all patients and analyzed their distribution and correlation. The patients were divided into three groups [Respiratory Syncytial Virus Immunoglobulin M (RSV-IgM) positive group A, Mycoplasma Immunoglobulin M (MP - IgM) positive group B, antibody - negative group with elevated hsCRP, 40 patients each]. We compared differences in hsCRP, platelet count, White Blood Cells (WBC), and Neutrophil (NE) among the groups.

Results: We conducted a systematic sorting and analysis of variables exhibiting significant differences. The results of the multivariate logistic regression analysis indicated that inflammatory markers, including white blood cell count (WBC) (OR 3.85, 95% CI: 1.116–1.623), neutrophils (NE) (OR 2.26, 95% CI: 1.091–1.312), high-sensitivity C-reactive protein (HsCRP) (OR 1.95, 95% CI: 1.068–14.640), lymphocytes (OR 1.30, 95% CI: 1.045–1.134), platelet count (OR 1.34, 95% CI: 1.625–2.760), and C-reactive protein (CRP) (OR 3.80, 95% CI: 1.232–2.379), were significantly associated with the presence of pathogenic bacteria.

Conclusion: There was significant correlation between inflammatory markers and pathogenic bacteria in patients with lower respiratory tract infection in Xinjiang region.

Keywords: correlation, inflammatory markers, lower respiratory tract infection, pathogenic bacteria

Introduction

Acute Lymphoblastic Leukemia Research Initiative (ALRI) is prevalent among children in China and endangers their health.¹ Lower respiratory tract infection is caused by pathogenic microorganisms invading and proliferating in the lower respiratory tract, including bronchitis and pneumonia.² This type of infection is generally attributed to bacteria, viruses, mycoplasma, fungi, and parasites, among other pathogens, and constitutes a prevalent infectious disease within the respiratory sector, particularly affecting pediatric populations.³ According to pertinent literature, approximately 2 million children under the age of five succumb to pneumonia each year worldwide, making it the foremost cause of child mortality in China.⁴ Consequently, the necessity for prompt and precise diagnostic measures, alongside targeted therapeutic interventions, is paramount.

In recent years, influenced by multiple factors such as the deterioration of the natural environment, the growth of population mobility, and climate change, the incidence of lower respiratory tract infection in children has exhibited an upward trend.⁵ Pathogens responsible for acute lower respiratory tract infection are predominantly categorized into three groups: bacteria, viruses, and atypical pathogens.⁶ With the extensive application of antibiotics, bacterial infections have declined, while viral and atypical pathogen infections have ascended.⁷ The nine - item joint screening for respiratory pathogens mainly comprises five viruses and four atypical pathogens. Researches have demonstrated that when children of different ages are afflicted with lower respiratory tract infections, there exist disparities in the degree of inflammatory response and the types of pathogens.⁸ Given that etiological test results are often not promptly obtainable, clinical treatment is mostly founded on empirical drug administration, which might lead to suboptimal therapeutic outcomes.⁹ The study examines nine pathogens, namely Respiratory Syncytial Virus, Influenza A Virus, Influenza B Virus, Parainfluenza Virus, Adenovirus, *Mycoplasma pneumoniae*, *Chlamydia pneumoniae*, *Legionella pneumophila* type 1, and *Rickettsia Q*, in the context of winter respiratory tract infections in Xinjiang. It considers the impact of climatic conditions, environmental factors, and population characteristics on the prevalence and transmission of respiratory tract infections in the region. In contrast to certain global or regional studies, the findings of this research more accurately represent the actual circumstances in Xinjiang and offer more specific guidance for clinical practice within this region. This study simultaneously detected nine common respiratory pathogens and analyzed their correlation with inflammatory markers, thereby offering a more comprehensive understanding of the etiology and condition of respiratory infections. Consequently, it provides a more precise foundation for clinical treatment. In light of the specificity of inflammatory markers induced by infections with different pathogens and the fact that these markers can be rapidly detected on the day of hospitalization or the subsequent day, the types of pathogens can be predicted by comparing the levels of inflammatory markers in children of different ages infected with different pathogens.¹⁰

In this study, indirect immunofluorescence (IFA) was employed to detect IgM antibodies of nine common respiratory pathogens, and simultaneously detect hypersensitive C - reactive protein and routine blood indexes. The aim was to analyze the prevalence of these pathogens in this region and their relationship with hsCRP, WBC, and neutrophil ratio, thereby identifying the main pathogens of lower respiratory tract infection in children and providing a scientific basis for rational clinical drug use.

Methods

Research Object

We strictly adhere to the STROBE checklist to ensure the transparency, reliability and validity of our research. Our study was approved by the Ethics Committee of Xinjiang Provincial People's Hospital (2019XE0130-3). A total of 120 patients diagnosed with lower respiratory tract infections were selected from the electronic medical records of Xinjiang Provincial People's Hospital, spanning the period from March 2019 to March 2023, for inclusion in a case-control study. In line with the principle of balanced clinicopathological features, the patients were divided into group A (RSV - IgM positive), group B (MP - IgM positive), and the antibody - negative group with elevated hsCRP, with 40 patients in each group. Clinicopathological data and follow - up information were complete for all patients. The screening process for the included patients is presented in [Figure 1](#).

Diagnostic Criteria

We included patients meeting the diagnostic criteria of the World Health Organization (WHO),¹¹ the American Academy of Pediatrics (AAP),¹² and the European Society of Infectious Diseases in Childhood (ESPID)¹³ for lower respiratory tract infections. Details are as follows:

Symptom observation: The patient presented with persistent cough, dyspnea, chest pain, fever, etc. Physical examination: Auscultation revealed moist rales, wheezing, or other abnormal lung breath sounds. Imaging studies, including chest X-rays and computed tomography (CT) scans, revealed pulmonary inflammatory regions characterized by alveolar infiltration, lung consolidation, or pleural effusion. Laboratory analyses indicated an elevated white blood cell count or increased neutrophil proportion, alongside heightened serum C-reactive protein (CRP) levels, an indicator of

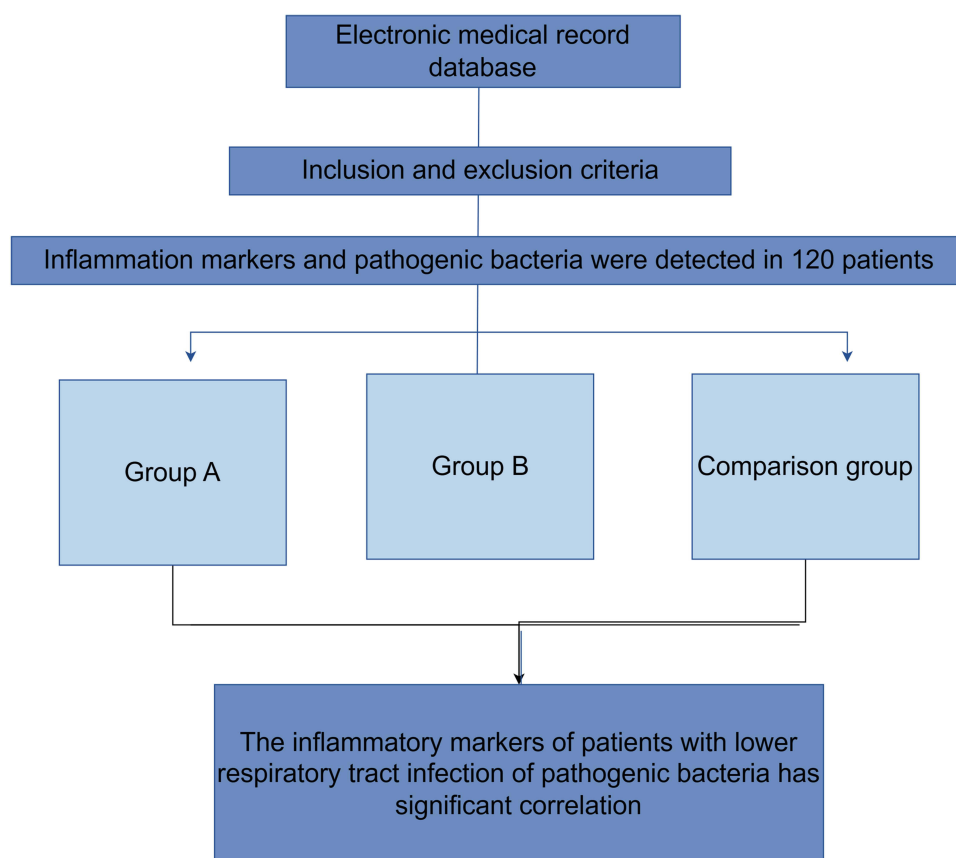


Figure 1 Included patient screening process (This figure was drawn by Figdraw. ID: PWIPA88040).

inflammation. Microbial investigations involved bacterial culture, viral antigen detection, or polymerase chain reaction (PCR) assays conducted on specimens such as nasopharyngeal swabs, sputum, and bronchial lavage fluid to ascertain the type of pathogen present.

Exclusion of Inclusion Criteria

In this study, the criteria for patient inclusion were delineated as follows: A diagnosis of lower respiratory tract infection was confirmed through clinical symptoms, laboratory test results, and imaging findings. The patient was required to be conscious and capable of actively participating in the research. Additionally, the patient's condition needed to be relatively stable, with no significant disease progression observed during the study period. Furthermore, it was essential that the patient's family members comprehensively understood the research objectives and provided informed consent.

In establishing the exclusion criteria for study participants, the following considerations are taken into account: individuals with other inflammatory diseases that could potentially confound laboratory test outcomes are excluded; those with systemic infections are also excluded; individuals with a history of pulmonary disease are not included; participants who have received antibiotic treatment within seven days prior to the study are excluded to mitigate any drug-related effects on the study results; and participants with incomplete or missing study data are also excluded. These criteria are aimed at maintaining the homogeneity of the study sample and the validity of the data.

Collection of Pathogenic Specimens

For each study participant, 2 mL of fasting venous blood was collected on the morning of the second day following hospitalization. The collected blood samples were then subjected to centrifugation at 3000 rpm for 10 minutes to achieve serum separation. The separated serum samples should be stored at a temperature of 2–8°C if analysis is to be conducted within 8 hours; if the analysis is delayed beyond 7 days, storage should be at –20°C. Additionally, on the day of

admission or the following day, peripheral blood samples were collected for routine hematological tests and high-sensitivity C-reactive protein (hs-CRP) analysis. For the purpose of etiological investigation, fluorescence microscopy was utilized to qualitatively assess IgM antibodies specific to nine pathogens present in serum samples. These pathogens included five respiratory viruses—respiratory syncytial virus, influenza A virus, influenza B virus, parainfluenza virus, and adenovirus—and four atypical pathogens: *Mycoplasma pneumoniae*, *Chlamydia pneumoniae*, *Legionella pneumophila* type 1, and *Rickettsia Q* thermal. This detection technique facilitates the effective identification and confirmation of pathogens responsible for lower respiratory tract infections. See Figure 2.

Pathogen Detection

Sample preparation: Serum samples are obtained from patients and stored under appropriate conditions.

Antigen plate preparation: Known respiratory viruses and atypical pathogens are immobilized on a slide to form an antigen plate.

Serum incubation: Dilute the serum sample and add it to the antigen plate, ensuring all fixed antigens are covered. Place the slide in a humid chamber and incubate at an appropriate temperature (usually 37°C) for a set period (usually 30 minutes to 1 hour).

Washing: After incubation, gently rinse the slide with PBS (phosphate - buffered saline) or a similar solution to remove unbound serum proteins.

Addition of fluorescently - labeled secondary antibodies: Add a fluorescently - labeled anti - human immunoglobulin (eg, FITC - labeled anti - human IgM) to each sample, followed by another incubation and washing.

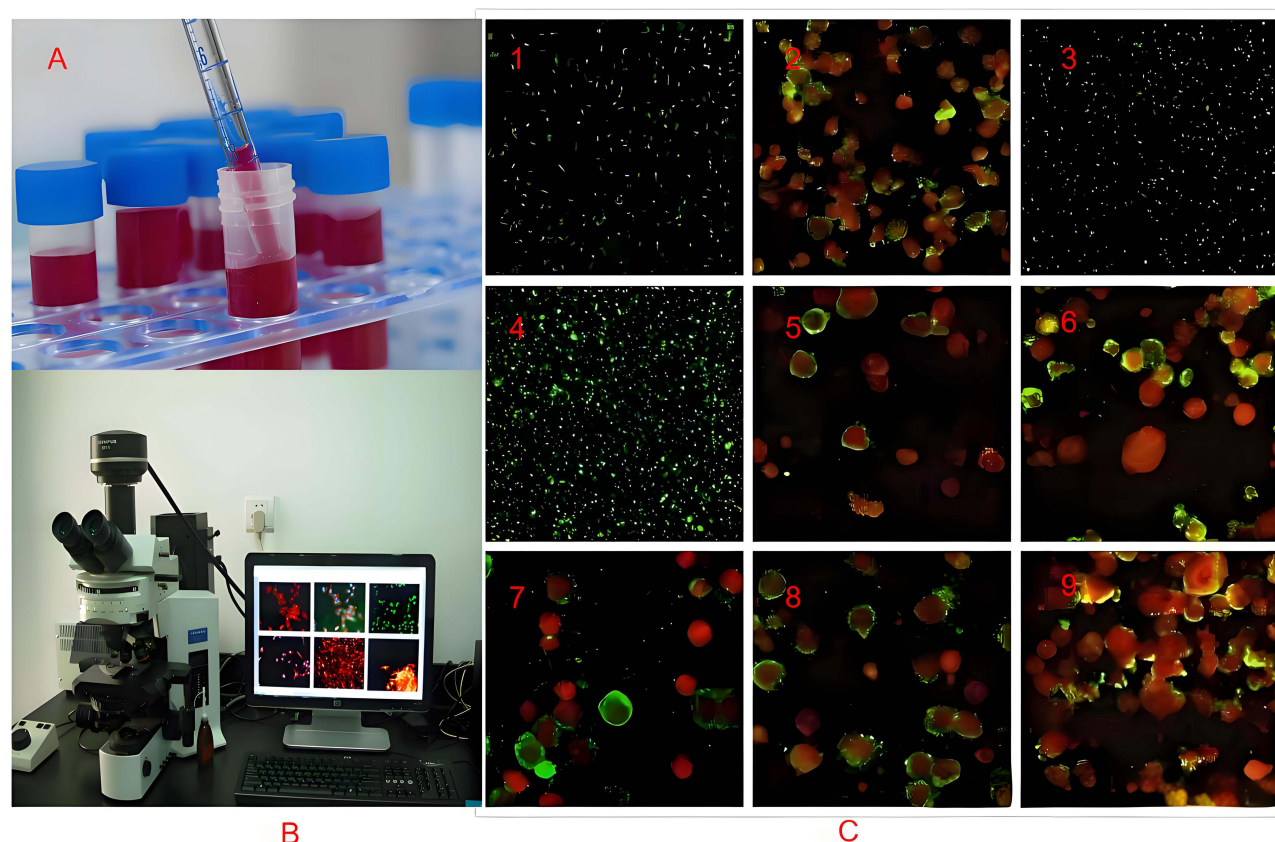


Figure 2 Fluorescence microscopy was used to detect nine pathogens in serum samples. (A) Serum sample (B) fluorescence microscope (C) (1) respiratory syncytial virus; (2) influenza A virus; (3) influenza B virus; (4) parainfluenza virus; (5) adenovirus; (6) *Mycoplasma pneumoniae*; (7) *Chlamydia pneumoniae*; (8) *Legionella pneumophila* type 1; (9) *Rickettsia Q* fever).

Fluorescence microscopy: Use a fluorescence microscope to observe the slide for specific fluorescence signals. If a bright fluorescence signal is detected in the region of a specific pathogen under the fluorescence microscope, it indicates the presence of specific IgM antibodies against that pathogen in the patient's serum. The intensity and distribution of the fluorescence signal can provide information about antibody titers and pathogen exposure.

This test encompasses the following respiratory viruses: *influenza A virus (IFVA)*, *respiratory syncytial virus (RSV)*, *influenza B virus (IFVB)*, *parainfluenza virus (PIV)*, and *adenovirus (ADV)*, as well as the following atypical pathogens: *Mycoplasma pneumoniae (MP)*, *Legionella pneumophila type 1 (LP1)*, *Chlamydia pneumoniae (CPN)*, and *Rickettsia Q thermondii (QFR)*. A positive result implies that the patient may have been recently infected with the corresponding pathogen, while a negative result may suggest no infection or antibody levels below the detection limit. See Figure 3.

Detection of Inflammatory Markers

Two milliliters of fasting venous blood is drawn from the patient and placed into a centrifuge tube. Subsequently, the serum is centrifuged at 3000 RPM for 10 minutes for isolation. In each reaction well of the slide, a diluted serum sample and a quality control product are added separately. The slide is placed in a humid chamber and incubated at a constant temperature of 37°C for 90 minutes to facilitate the binding of antibodies in the serum to the immobilized antigen. After incubation, the slides are washed twice with PBS (phosphate buffer) and once with distilled water to remove unbound serum components and impurities, and then air-dried to prepare for the subsequent steps. A luciferin-labeled secondary antibody is added to each reaction well of the slide, which is then placed in a humid chamber and incubated at 37°C for 30 minutes to allow the luciferin-labeled secondary antibody to react with the bound antibody. After incubation, the washing steps with PBS and distilled water are repeated to ensure the removal of unbound fluorescein-labeled

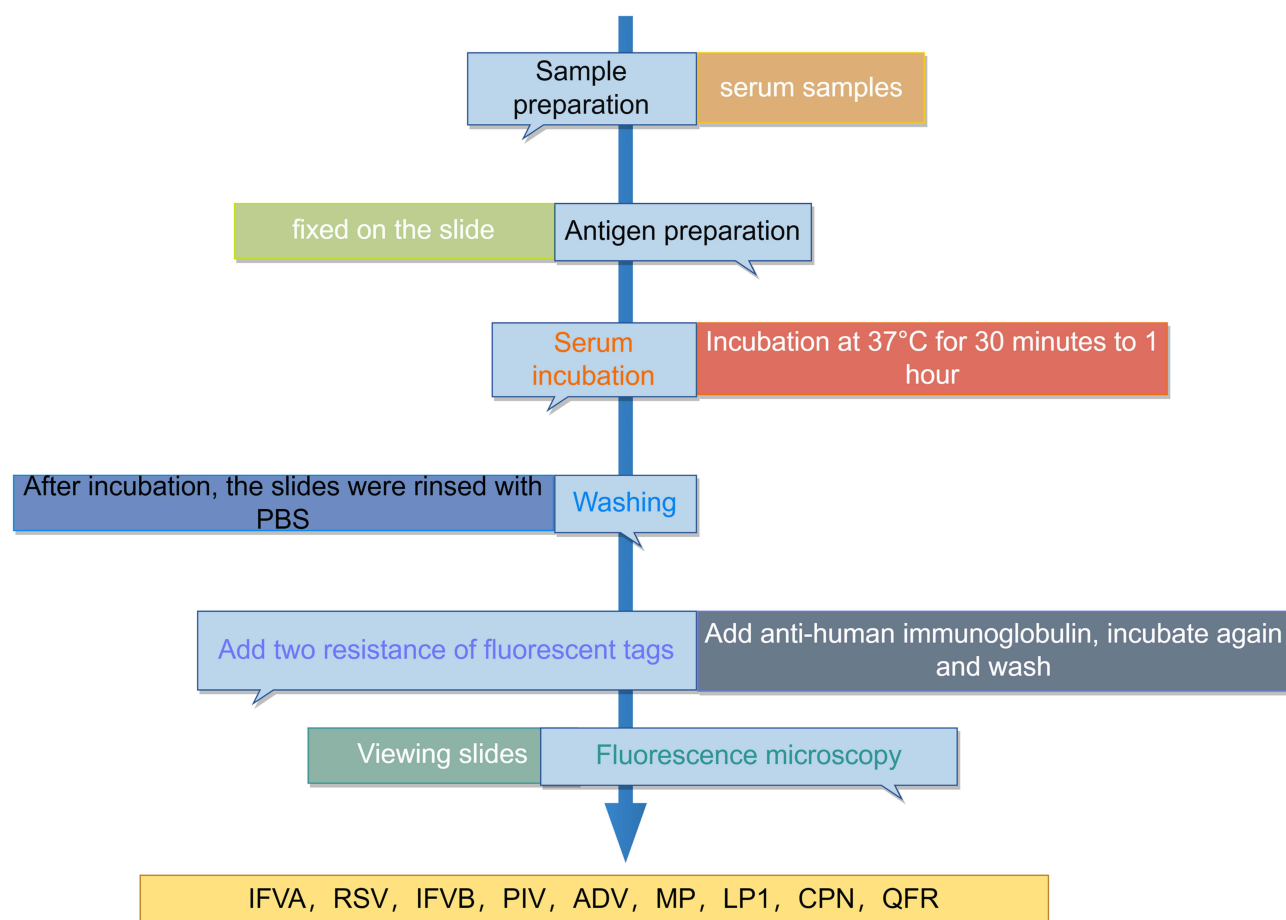


Figure 3 Flow chart of pathogen detection. (This image was created by Figdraw. ID: RPAPP97379).

secondary antibodies. The slide is air - dried again. A drop of mounting medium is added to each reaction well of the slide, and then a coverslip is placed over it. Finally, the specific fluorescence signal is observed and recorded under a fluorescence microscope at a 400 - fold magnification.

Detection of Hypersensitive C-Reactive Protein

Firstly, retrieve the reaction plate and detection buffer required for the experiment. Place them at room temperature and allow 10 minutes for equilibration. Verify that the ID chip has been correctly inserted into the instrument and ensure that the batch number of the reaction plate corresponds to that of the ID chip to ensure the accuracy of the experimental data. Employ a sampler to puncture the foil on the detection buffer tube and draw 10 microliters of the peripheral blood sample. Wipe the sampler to eliminate blood samples from the periphery of the capillaries so as to avoid external contamination. Add the processed blood sample to the buffer tube, then seal the tube and mix thoroughly. Repeat this operation five times to ensure the complete mixing of the blood sample with the buffer. Open the lid of the tube, dispense two drops of the mixture, and subsequently add two drops of the sample mixture to the loading hole of the reaction plate. Place the reaction plate at room temperature for three minutes. Insert the reaction plate into the sample rack of the i - CHROMA Reader immunofluorescence analyzer and press the “SELECT” key to initiate scanning. Ensure that the reaction plate is correctly oriented and fully inserted into the instrument. The instrument will automatically commence scanning the reaction plate and display the scan results on the display screen of the i - CHROMA Reader immunofluorescence analyzer.

Sample Result Judgment

To ensure the accuracy of the experiment and the validity of the kit, positive and negative control groups were established for each experiment. The reliability of the experimental procedure and the kit can be verified through the observation of these control groups' results.

Under a fluorescence microscope, the positive control group is expected to present a specific fluorescence pattern. Specifically, adenoviruses, influenza viruses, respiratory syncytial viruses, or parainfluenza viruses should exhibit apple - green fluorescence in the nucleus, cytoplasm, or membrane within 1–15% of positive - control cells. Syncytial phenomena may also be observed in parainfluenza virus and respiratory syncytial virus. All bacteria within Legionella, Chlamydia, or Rickettsiella should display apple - green fluorescence. Mycoplasma should show apple - green fluorescence at the periphery of positive - quality - control cells.

In the negative control group, Legionella, Chlamydia, or Rickettsiella should not fluoresce, while cells of mycoplasma, adenovirus, influenza A and B viruses, respiratory syncytial virus, and parainfluenza viruses should appear red.

For positive results, apple - green fluorescence in the nucleus, cytoplasm, or membrane of respiratory syncytial virus, influenza virus, adenovirus, or parainfluenza virus is anticipated to be observed in 1–15% of cells with positive sera. All bacteria within Legionella, Chlamydia, or Rickettsiella should exhibit apple - green fluorescence. Mycoplasma should display apple - green fluorescence at the periphery of cells sensitive to positive serum.

For negative results, no fluorescence is expected to be observed for Legionella, Chlamydia pneumoniae, or Rickettsiae, while cells of mycoplasma, adenovirus, influenza A and B viruses, respiratory syncytial virus, and parainfluenza viruses should appear red.

HsCRP normal value <0.5 mg/L. The normal value of white blood cells is $(4\sim10)\times10^9/L$; Neutrophils are normal. $N<40\%$ (less than 1 year old), $N<50\%$ (1~4 years old), $N\approx50\%$ (4~6 years old), $N<65\%$ (more than 6 years old).

Statistical Analysis

The data were imported into Statistic Package for Social Science (SPSS) 26.0 software (IBM, Armonk, NY, USA) for statistical analysis. Counting data were expressed as integers or percentages, chi-square tests (χ^2 -tests) were used for comparison between groups, and rank-sum tests were used for ordered variables. Measurement data were expressed as mean \pm standard deviation. For the data meeting the normal distribution, the two independent samples *t* test is used to compare. If the data did not meet the normal distribution, the Mann–Whitney U rank sum test was used. Single factor analysis of variance was used for the comparison of multiple groups of quantitative data, and SNK-q test was used for

pair comparison among multiple groups. Ordered multiple Logistic regression was used to analyze the correlation between inflammatory markers and pathogenic bacteria. $P_{\text{value}} < 0.05$ was set as a statistically significant difference.

Results

Univariate Analysis of Clinical Data

In groups A, B, and the control group, the ages were 6.6 ± 2.8 , 6.7 ± 1.6 , and 6.9 ± 1.8 years respectively ($F = 0.03$, $P = 0.994$), male proportions were 57.50%, 45.00%, and 42.00% ($X^2 = 0.51$, $P = 0.916$), smoking history proportions were 42.00%, 40.00%, and 35.00% ($X^2 = 0.77$, $P = 0.858$). In groups A and B, disease durations were 16.15 ± 4.98 and 15.77 ± 4.63 months ($t = 0.17$, $P = 0.845$). BMIs were 22.76 ± 2.85 , 22.50 ± 2.92 , and 22.31 ± 2.64 ($F = 0.18$, $P = 0.910$), systolic blood pressures were 116.46 ± 10.75 , 117.49 ± 12.03 , and 118.52 ± 11.44 ($F = 0.25$, $P = 0.864$), diastolic blood pressures were 78.48 ± 4.79 , 78.95 ± 4.86 , and 79.04 ± 5.10 ($F = 0.19$, $P = 0.902$), TC levels were 3.64 ± 0.98 , 3.42 ± 0.95 , and 3.49 ± 1.13 ($F = 0.55$, $P = 0.652$), TG levels were 1.26 ± 0.42 , 1.22 ± 0.43 , and 1.25 ± 0.41 ($F = 0.20$, $P = 0.898$), HDL - C levels were 1.34 ± 0.25 , 1.29 ± 0.31 , and 1.33 ± 0.32 ($F = 0.20$, $P = 0.897$), LDL - C levels were 2.26 ± 0.52 , 2.29 ± 0.59 , and 2.34 ± 0.58 ($F = 0.11$, $P = 0.954$), with no significant differences among groups for all these parameters. See Table 1.

Pathogen Distribution and Anti-Nuclear Antibody Positive Statistics

In Group A, B, and the control group, the proportions of simple virus infection were 30.0%, 32.5%, and 27.5% respectively, with no significant difference ($X^2 = 0.238$, $P = 0.888$). For *Mycoplasma pneumoniae* infection, the proportions in the three groups were 25.0%, 27.5%, and 32.5% respectively, showing no significant difference ($X^2 = 0.575$, $P = 0.750$). The proportions of simple bacterial infection in the three groups were 32.5%, 30.0%, and 30.0% respectively, with no statistically significant difference ($X^2 = 0.078$, $P = 0.962$). Regarding co - infection, the proportions in the three groups were 37.5%, 45.0%, and 37.5% respectively, and there was no statistically significant difference ($X^2 = 0.625$, $P = 0.732$). See Table 2 and Figure 4.

Table 1 Univariate Analysis of Clinical Data Between the Two Groups ($\bar{x} \pm \text{sd}$, (n(%)))

	Group A (40)	Group B (40)	Comparison group (40)	F/t/ χ^2	P-value
Age (years)	6.6 ± 2.8	6.7 ± 1.6	6.9 ± 1.8	0.03	0.994
Gender (Male)	23(57.50)	18(45.00)	17(42.00)	0.51	0.916
Smoking history	17(42.00)	16(40.00)	14(35.00)	0.77	0.858
Sick time (month)		16.15 ± 4.98	15.77 ± 4.63	0.17	0.845
BMI (kg/m^2)	22.76 ± 2.85	22.50 ± 2.92	22.31 ± 2.64	0.18	0.910
Systolic pressure (mmHg)	116.46 ± 10.75	117.49 ± 12.03	118.52 ± 11.44	0.25	0.864
Diastolic blood pressure (mmHg)	78.48 ± 4.79	78.95 ± 4.86	79.04 ± 5.10	0.19	0.902
TC (mmol/L)	3.64 ± 0.98	3.42 ± 0.95	3.49 ± 1.13	0.55	0.652
TG (mmol/L)	1.26 ± 0.42	1.22 ± 0.43	1.25 ± 0.41	0.20	0.898
HDL-C (mmol/L)	1.34 ± 0.25	1.29 ± 0.31	1.33 ± 0.32	0.20	0.897
LDL-C (mmol/L)	2.26 ± 0.52	2.29 ± 0.59	2.34 ± 0.58	0.11	0.954

Table 2 Pathogen Distribution Statistics of Patients in the Two Groups (n(%))

	Group A (40)	Group B (40)	Comparison group (40)	χ^2	P-value
Simple virus infection	12 (30.0)	13 (32.5)	11 (27.5)	0.238	0.888
Mycoplasma pneumoniae infection	10 (25.0)	11 (27.5)	13 (32.5)	0.575	0.750
Simple bacterial infection	13 (32.5)	12 (30.0)	12 (30.0)	0.078	0.962
Mixed infection	15 (37.5)	18 (45.0)	15 (37.5)	0.625	0.732

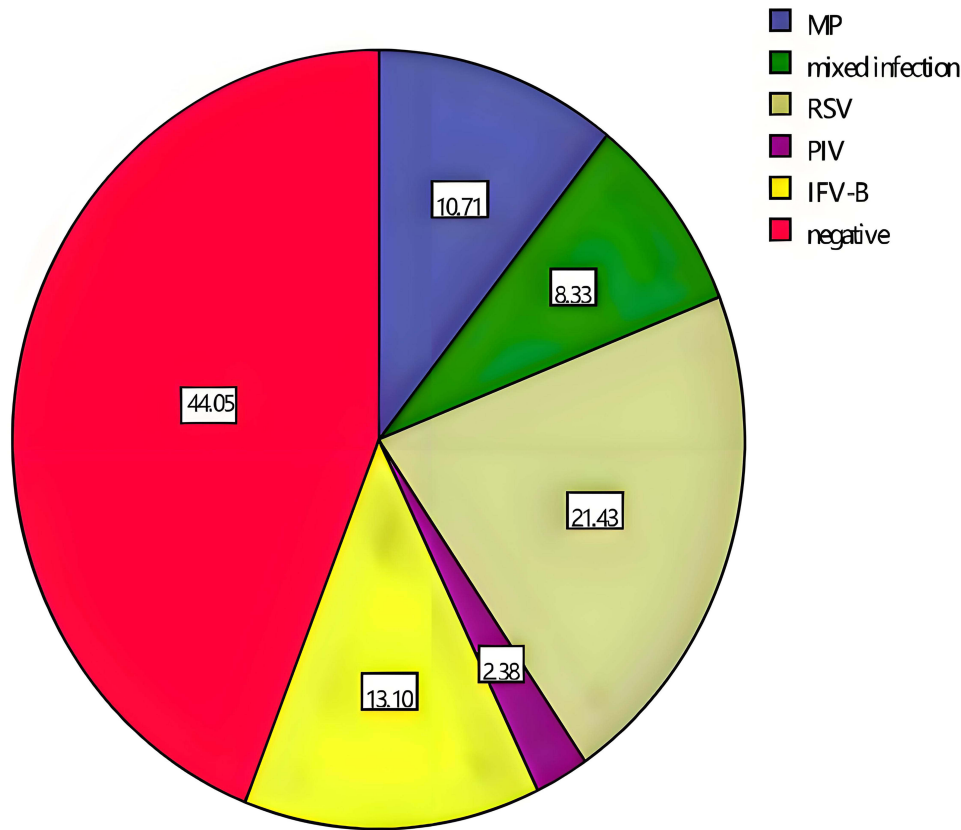


Figure 4 Statistics of positive anti-nuclear antibody.

Comparison of Blood Routine and Blood HsCRP

In Group A, the average white blood cell count was 9.8 ± 2.1 ; in Group B, it was 6.6 ± 1.6 ; and in the control group, it was 14.7 ± 3.1 . The difference in white blood cell counts among the three groups was statistically significant ($F = 120.073$, $P < 0.0001$). The neutrophil percentages in Group A and Group B were $61.9 \pm 6.5\%$ and $59.5 \pm 7.8\%$ respectively, while in the control group it was $83.5 \pm 5.1\%$. The difference in neutrophil percentages among the three groups was statistically significant ($F = 162.404$, $P < 0.0001$). The average Hs - CRP levels were 28.2 ± 5.1 mg/L in Group A and 25.6 ± 3.1 mg/L in Group B, compared with 37.1 ± 5.8 mg/L in the control group. The difference in Hs - CRP levels among the three groups was statistically significant ($F = 63.014$, $P < 0.0001$). See Table 3.

Comparison of Inflammatory Markers of Different Pathogens

In Group A and Group B, the mean lymphocyte counts were 1.49 ± 0.13 and 1.20 ± 0.11 respectively, compared with 1.82 ± 0.15 in the control group. The difference in lymphocyte counts among the three groups was statistically significant ($F = 224.233$, $P < 0.0001$). The average platelet counts in Group A and Group B were 0.80 ± 0.06 and 0.69 ± 0.05 respectively, compared with

Table 3 Comparison of Blood Routine and Blood HsCRP Between the Two Groups ($\bar{x} \pm \text{sd}$)

	Group A (40)	Group B (40)	Comparison group (40)	F	P-value
WBC ($\times 10^9/\text{L}$)	9.8 ± 2.1	6.6 ± 1.6	14.7 ± 3.1	120.073	<0.0001
NE (%)	61.9 ± 6.5	59.5 ± 7.8	83.5 ± 5.1	162.404	<0.0001
Hs-CRP (mg/l)	28.2 ± 5.1	25.6 ± 3.1	37.1 ± 5.8	63.014	<0.0001

Table 4 Comparison of Inflammatory Markers of Different Pathogens ($\bar{x} \pm s$)

	Group A (40)	Group B (40)	Comparison group (40)	F	P-value
Lymphocyte ($\times 10^9/L$)	1.49 \pm 0.13	1.20 \pm 0.11	1.82 \pm 0.15	224.233	<0.0001
Platelet count ($\times 10^9/L$)	0.80 \pm 0.06	0.69 \pm 0.05	0.87 \pm 0.06	101.855	<0.0001
CRP (mg/dL)	219.72 \pm 22.41	159.33 \pm 18.64	253.41 \pm 24.85	185.839	<0.0001
Granulocyte (%)	25.88 \pm 2.71	16.23 \pm 1.84	33.31 \pm 2.90	459.833	<0.0001

Table 5 Logistic Multivariate Analysis of Inflammatory Markers and Pathogens

	β	S.E.	Wald χ^2	OR (95% CI)	P-value
WBC ($\times 10^9/L$)	0.658	0.213	9.535	3.85(1.116~1.623)	0.007
NE (%)	0.198	0.057	12.298	2.26(1.091~1.312)	0.032
HsCRP (mg/l)	0.101	0.175	10.336	1.95(1.068~14.640)	0.024
Lymphocyte ($\times 10^9/L$)	0.267	0.165	12.606	1.30(1.045~1.134)	0.015
Platelet count ($\times 10^9/L$)	0.611	0.119	19.450	1.34(1.625~2.760)	0.005
CRP (mg/dL)	0.374	3.277	21.825	3.80(1.232~2.379)	<0.0001
Granulocyte (%)	0.045	2.109	3.662	8.64(1.467~2.865)	0.097

0.87 \pm 0.06 in the control group. The difference in platelet counts among the three groups was statistically significant ($F = 101.855$, $P < 0.0001$). In Group A, the average CRP level was 219.72 \pm 22.41 mg/dL; in Group B, it was 159.33 \pm 18.64 mg/dL; and in the control group, it was 253.41 \pm 24.85 mg/dL. The difference in CRP levels among the three groups was statistically significant ($F = 185.839$, $P < 0.0001$). In Group A, the granulocyte percentage was 25.88 \pm 2.71%; in Group B, it was 16.23 \pm 1.84%; and in the control group, it was 33.31 \pm 2.90%. The difference in granulocyte percentage among the three groups was statistically significant ($F = 459.833$, $P < 0.0001$). See Table 4.

Correlation Between Inflammatory Markers and Pathogenic Bacteria

We examined variables with significant differences for sorting and analysis. Through multivariate Logistic analysis, we found that WBC (OR(95% CI)=3.85(1.116~1.623), NE (OR(95% CI)=2.26(1.091~1.312), HsCRP (OR (95% CI) = 1.95 (1.068 ~ 14.640), lymphocytes (OR (95% CI) = 1.30 (1.045 ~ 1.134). Platelet count (OR(95% CI)=1.34(1.625~2.760) and CRP(OR(95% CI)=3.80(1.232~2.379) and other inflammatory markers were correlated with pathogenic bacteria. See Table 5.

Discussion

Lower respiratory tract infection seriously threatens children's health, growth, and development. Once it occurs, the condition usually deteriorates rapidly and has a high risk of complications.¹⁴ Consequently, early and accurate diagnosis of pathogen types in patients and targeted treatment measures are crucial for shortening the recovery time.¹⁵ Currently, the results of etiological testing often take some time to be obtained. In emergency situations, clinical treatment often depends on empirical medication to timely control the disease.¹⁶ This may lead to the irrational use of antibiotics, which not only prolongs the patient's recovery time but also may increase the risk of treatment failure, ultimately affecting the patient's prognosis.

This study's results showed that hsCRP levels in RSV - IgM - positive children were slightly elevated compared to normal and significantly different from the control group, consistent with other research and viral infection characteristics.¹⁷ Serum CRP, an acute - phase reactant protein produced during body injury or inflammation, can be detected in acute - phase patients with infectious and non - infectious diseases, thus being a non - specific reactant to tissue injury.¹⁸ In normal blood, CRP is low. But with inflammatory diseases and tissue damage, the liver synthesizes large amounts of CRP. Blood CRP levels start to rise within 6-8 hours, peak within 24-72 hours (up to 1000 times normal), and decline significantly after 24 - hour anti - infection

treatment.¹⁹ In viral infection, intact cell membranes prevent phospholipid protein exposure, so CRP production is not triggered and blood CRP concentration does not increase significantly.²⁰

HsCRP, with high detection sensitivity, can distinguish bacterial and viral infections and has been widely used clinically in recent years.²¹ With the increasing rate of viral respiratory infections in children, hsCRP's diagnostic value in pediatric infectious diseases has become more important.²² Dynamic hsCRP detection is crucial when other hematological findings are unclear or obscured, such as in granulocytopenia, deficiency, or immunosuppressant use.²³ Clinically, HsCRP levels are often used to assess inflammation extent and infection likelihood.²⁴ HsCRP > 10 mg/L may indicate other infections; 10–20 mg/L may suggest viral or mild bacterial infection; 20–50 mg/L may indicate general bacterial infection. Due to lack of exposed phospholipid proteins on cell membranes during viral infection, serum CRP concentration does not change much.

In this study, antibody - negative patients had significantly higher WBC levels, NE proportions, and Hs - CRP levels than other groups, while RSV - antibody - negative patients had significantly lower levels. This shows different inflammation levels in lower respiratory tract infections caused by antibody - negative agents, and a non - obvious inflammatory response in RSV - only infections. In triple - virus - co - infected patients with RSV, serum HsCRP was > 10 mg/mL, but in RSV - only - infected patients, it was < 10 mg/L, suggesting RSV may cause more obvious inflammation when combined with other viruses.

The WBC - to - NE ratio is important for infection diagnosis. In bacterial infections, this ratio usually rises significantly.²⁵ For patients with low WBC baselines, even a 1 - to - several - fold increase may be within normal range, so WBC has low sensitivity and specificity.²⁶ About 25.9% of children with elevated WBC or neutrophil counts have normal hsCRP levels.²⁷ Thus, for infection diagnosis, hsCRP and WBC count or neutrophil ratio should be measured together to avoid misdiagnosis. Dynamic monitoring of these indicators is crucial for rational antibiotic use, treatment - effect observation, and drug - withdrawal indication.²⁸

Elevated WBC is commonly due to inflammation and acute infection, and the increase degree is related to pathogen type, infection site, and individual differences.²⁹ Bacterial infections cause the most pronounced WBC increase. In respiratory virus infections, a child's WBC may be normal or slightly decreased. Due to large individual differences in children's WBC baselines and wide normal ranges, at disease onset, WBC may increase only 1 - several times, and some low - baseline children may have normal total WBC even during bacterial infection. Dynamic monitoring of patients' WBC, NE, and HsCRP levels can help judge disease changes and guide rational drug use.³⁰

This study enhances the comprehension of regional variations in respiratory tract infections by examining winter respiratory tract infections in Xinjiang. Furthermore, the findings align with the contemporary focus on precision medicine, which aims to offer patients personalized treatment plans through a more precise understanding of disease mechanisms and associated factors. Notably, the study observed elevated white blood cell (WBC) levels in patients with antibody-negative infections, whereas patients with isolated respiratory syncytial virus (RSV) infections exhibited comparatively lower WBC levels. This indicates that leukocyte count may serve as an auxiliary marker in differentiating various types of respiratory infections within clinical diagnostics. In patients presenting with unexplained fever and respiratory symptoms, a marked elevation in leukocyte count may suggest the presence of antibody-negative infectious agents, necessitating further investigation for additional pathogens. Conversely, if the leukocyte count is normal or only marginally elevated, there is a higher likelihood of respiratory syncytial virus (RSV) infection; however, this assessment should be integrated with other diagnostic indicators to ensure a comprehensive evaluation. The prevalence of neutrophils (NE) was observed to be elevated in patients with antibody-negative infections, whereas it was comparatively lower in individuals with uncomplicated respiratory syncytial virus (RSV) infections. As a critical component of leukocytes, the proportion of NE holds significant clinical relevance. An increased NE ratio may suggest the presence of bacterial infections or other inflammatory stimuli. In patients with antibody-negative infections, this elevation could indicate the presence of undetected pathogens or a heightened inflammatory response. In clinical practice, this observation may indicate the necessity for a more proactive investigation into the underlying causes and the consideration of antibiotic therapy. However, such decisions must be made judiciously to prevent the inappropriate use of antibiotics. The HsCRP levels were elevated in patients with antibody-negative infections and comparatively lower in those with uncomplicated RSV infections. As a sensitive marker of inflammation, variations in HsCRP levels can assist clinicians in discerning the type of infection and evaluating the magnitude of the inflammatory response. Elevated levels of high-sensitivity

C-reactive protein (HsCRP) may suggest the presence of bacterial infections or other conditions characterized by a pronounced inflammatory response. In contrast, HsCRP levels typically remain unremarkable in uncomplicated respiratory syncytial virus (RSV) infections, reflecting the nature of viral pathogenesis. Consequently, HsCRP levels serve as a crucial diagnostic marker in clinical practice for differentiating between viral and bacterial infections, thereby playing a significant role in informing appropriate therapeutic interventions.

This study has several implementation limitations. Limited by budget and time, the sample size was small, potentially lacking statistical power to represent the whole population. The sample - examination time span (< 1 year) restricted the study of seasonal - change impacts on test results as seasonal factors can affect pathogen prevalence, and the sample size was insufficient to capture these. Some patients' families did not cooperate with blood collection, and some patients discharged early for economic reasons, preventing continuous monitoring, which restricts tracking of disease progression and treatment effects and affects prognosis. Previous local treatment for some patients might have affected results, so the experimental results may not fully reflect the untreated state. Non - standard experimental operations may cause cross - contamination during sample addition or washing, leading to false - positive results. This needs to be resolved by improving the operation process and operator training.

This study is subject to several limitations. Variables such as climate, environmental conditions, population genetic background, and pathogen prevalence across different regions may influence the relationship between inflammatory markers and pathogenic bacteria. Certain regions may facilitate the survival and dissemination of specific pathogens due to unique climatic conditions, leading to variations in infection types and inflammatory responses. Consequently, this study further investigates the correlation between inflammatory markers and pathogenic bacteria. The development and functionality of the immune system vary across different age groups, leading to diverse immune responses to pathogens. Consequently, when extrapolating the findings of this study to other age demographics, adjustments and verifications are conducted in accordance with the specific characteristics of each age group to ensure relevance and applicability. Furthermore, healthcare settings may exhibit variability in testing methodologies, diagnostic criteria, and treatment protocols. Therefore, the implementation of this research across diverse healthcare environments is designed to ensure that the findings are appropriately adapted and applied within various clinical contexts.

In future research endeavors, it is imperative to enhance the sample size by engaging in multi-center collaborations, specifically by partnering with other medical institutions within the Xinjiang region and establishing joint research initiatives with medical entities in other regions of China that exhibit analogous climatic or disease characteristics. Such collaborations will facilitate the collection of a more diverse array of case types, thereby augmenting the representativeness and generalizability of the research findings. Additionally, it is essential to extend the temporal scope of the study to encompass multiple seasons or even several years. This approach allows for a more comprehensive observation of the seasonal variations in respiratory infections and the potential seasonal differences in the relationship between inflammatory markers and pathogenic bacteria. It is imperative to conduct an in-depth analysis of the mechanisms by which seasonal factors influence respiratory tract infections. Future research should explore various dimensions, including climatic conditions, population behaviors, and the survival and transmission characteristics of pathogens, to elucidate the impact of seasonal variations on research outcomes.

Conclusion

This study demonstrated that critical inflammatory markers in patients' blood, including white blood cell count, neutrophil percentage, high-sensitivity C-reactive protein (Hs-CRP), lymphocyte count, platelet count, C-reactive protein (CRP), and granulocyte percentage, are significantly associated with the type of pathogenic bacteria and the severity of infection. However, limitations such as the small sample size, the brief duration of sample examination, and the lack of cooperation from some patients may affect the generalizability and accuracy of the findings. In future studies, the sample size should be expanded, the study time span extended, and the molecular mechanism of the interaction between inflammatory markers and pathogenic bacteria should be further explored, so as to provide reference for subsequent studies.

Ethics Approval and Consent to Participate

This study was approved by the Ethical Committee of Traditional Chinese Medicine Hospital Affiliated to Xinjiang Medical University, batch number: 20190129. Signed written informed consents were obtained from the patients and/or guardians. This study was conducted in accordance with the Declaration of Helsinki and followed the ethical standards of China.

Funding

This work was supported by the Study on the mechanism of SangxingZhike granule in regulating fluid metabolism disorder and inhibiting ferroptosis in cough variant asthma based on the theory of “lung Tong Tiaoshui” 2022D01E80.

Disclosure

The authors declared no conflicts of interest in this work.

References

1. Binks MJ, Beissbarth J, Oguoma VM, et al. Acute lower respiratory infections in Indigenous infants in Australia's Northern Territory across three eras of pneumococcal conjugate vaccine use (2006-15): a population-based cohort study. *Lancet Child Adolesc.* 2020;4(6):425–434. doi:10.1016/S2352-4642(20)30090-0
2. Cheng J, Su H, Xu Z. Intraday effects of outdoor air pollution on acute upper and lower respiratory infections in Australian children. *Environ Pollut.* 2021;268(Pt A):115698. doi:10.1016/j.envpol.2020.115698
3. Annamalai AA, Abbott S, Khoo SK, et al. The impact of cytokine levels in young South African children with and without HIV-associated acute lower respiratory infections. *J Med Virol.* 2021;93(6):3647–3655. doi:10.1002/jmv.26730
4. Wang X, Li Y, Mei X, et al. Global hospital admissions and in-hospital mortality associated with all-cause and virus-specific acute lower respiratory infections in children and adolescents aged 5-19 years between 1995 and 2019: a systematic review and modelling study. *BMJ Glob Health.* 2021;6(7):e006014. doi:10.1136/bmjgh-2021-006014
5. Bleakley AS, Licciardi PV, Binks MJ. Vitamin D modulation of the innate immune response to paediatric respiratory pathogens associated with acute lower respiratory infections. *Nutrients.* 2021;13(1):276. doi:10.3390/nut13010276
6. Wang X, Li Y, O'Brien KL, et al. Global burden of respiratory infections associated with seasonal influenza in children under 5 years in 2018: a systematic review and modelling study. *Lancet Glob Health.* 2020;8(4):e497–e510. doi:10.1016/S2214-109X(19)30545-5
7. Chu FL, Li C, Chen L, et al. Respiratory viruses among pediatric inpatients with acute lower respiratory tract infections in Jinan, China, 2016-2019. *J Med Virol.* 2022;94(9):4319–4328. doi:10.1002/jmv.27875
8. Heidemann J, Konrad M, Roderburg C, et al. Association between antihypertensive therapy and risk of acute lower respiratory infections (ALRI): a retrospective cohort study. *Healthcare-Basel.* 2022;10(11):2318. doi:10.3390/healthcare10112318
9. Abudureyimu K, Suryadhi M, Yorifuji T, et al. Exposure to fine particulate matter and acute upper- and lower-respiratory tract infections (AURI and ALRI) in children under five years of age in India. *Arch Environ Occup H.* 2023;78(1):1–6. doi:10.1080/19338244.2022.2047584
10. Kubale J, Kujawski S, Chen I, et al. Etiology of acute lower respiratory illness hospitalizations among infants in 4 countries. *Open Forum Infect Di.* 2023;10(12):ofad580. doi:10.1093/ofid/ofad580
11. Klink T, Rankin DA, Piya B, et al. Evaluating the diagnostic accuracy of the WHO severe acute respiratory infection (SARI) criteria in Middle Eastern children under two years over three respiratory seasons. *PLoS One.* 2020;15(4):e0232188. doi:10.1371/journal.pone.0232188
12. Chan K, Sisk B, Yun K, et al. Global health experience and interest: results from the AAP periodic survey. *Pediatrics.* 2020;145(1). doi:10.1542/peds.2019-1655
13. Dornbusch HJ, Vesikari T, Guarino A, et al. Rotavirus vaccination for all children or subgroups only? Comment of the European Academy of Paediatrics (EAP) and the European society for paediatric infectious diseases (ESPID) recommendation group for rotavirus vaccination. *Eur J Pediatr.* 2020;179(9):1489–1493. doi:10.1007/s00431-020-03608-5
14. Wang X, Li Y, Shi T, et al. Global disease burden of and risk factors for acute lower respiratory infections caused by respiratory syncytial virus in preterm infants and young children in 2019: a systematic review and meta-analysis of aggregated and individual participant data. *Lancet.* 2024;403(10433):1241–1253. doi:10.1016/S0140-6736(24)00138-7
15. Wang X, Li Y, Deloria-Knoll M, et al. Global burden of acute lower respiratory infection associated with human metapneumovirus in children under 5 years in 2018: a systematic review and modelling study. *Lancet Glob Health.* 2021;9(1):e33–e43. doi:10.1016/S2214-109X(20)30393-4
16. Broor S, Parveen S, Maheshwari M. Respiratory syncytial virus infections in India: epidemiology and need for vaccine. *Indian J Med Microbi.* 2018;36(4):458–464. doi:10.4103/ijmm.IJMM_19_5
17. Smedemark SA, Aabenhus R, Llor C, et al. Biomarkers as point-of-care tests to guide prescription of antibiotics in people with acute respiratory infections in primary care. *Cochrane Db Syst Rev.* 2022;10(10):CD010130. doi:10.1002/14651858.CD010130.pub3
18. Yong SJ, Halim A, Halim M, et al. Inflammatory and vascular biomarkers in post-COVID-19 syndrome: a systematic review and meta-analysis of over 20 biomarkers. *Rev Med Virol.* 2023;33(2):e2424. doi:10.1002/rmv.2424
19. Yin H, Mo S. Value of combined detection of serum amyloid A, C-reactive protein and procalcitonin in differential diagnosis of respiratory tract infection in children of China. *Ann Med.* 2022;54(1):1732–1737. doi:10.1080/07853890.2022.2064542
20. Carbonell R, Urgeles S, Salgado M, et al. Negative predictive value of procalcitonin to rule out bacterial respiratory co-infection in critical covid-19 patients. *J Infection.* 2022;85(4):374–381. doi:10.1016/j.jinf.2022.06.024
21. Lindstrom A, Tangden T. Introducing the C-reactive protein point-of-care test: a conversation analytic study of primary care consultations for respiratory tract infection. *Soc Sci Med.* 2022;315:115493. doi:10.1016/j.socscimed.2022.115493

22. Thornton CS, Caverly LJ, Kalikin LM, et al. Prevalence and clinical impact of respiratory viral infections from the STOP2 study of cystic fibrosis pulmonary exacerbations. *Ann Am Thorac Soc*. 2024;21(4):595–603. doi:10.1513/AnnalsATS.202306-576OC
23. Plebani M. Why C-reactive protein is one of the most requested tests in clinical laboratories? *Clin Chem Lab Med*. 2023;61(9):1540–1545. doi:10.1515/cclm-2023-0086
24. Li Y, Min L, Zhang X. Usefulness of procalcitonin (PCT), C-reactive protein (CRP), and white blood cell (WBC) levels in the differential diagnosis of acute bacterial, viral, and mycoplasmal respiratory tract infections in children. *Bmc Pulm Med*. 2021;21(1):386. doi:10.1186/s12890-021-01756-4
25. Wang XX, Ma CT, Jiang YX, et al. Cefoperazone sodium/sulbactam sodium vs piperacillin sodium/tazobactam sodium for treatment of respiratory tract infection in elderly patients. *World J Clin Cases*. 2021;9(29):8694–8701. doi:10.12998/wjcc.v9.i29.8694
26. Downey DG, Brockbank S, Martin SL, et al. The effect of treatment of cystic fibrosis pulmonary exacerbations on airways and systemic inflammation. *Pediatr Pulm*. 2007;42(8):729–735. doi:10.1002/ppul.20646
27. Davies PL, Spiller OB, Beeton ML, et al. Relationship of proteinases and proteinase inhibitors with microbial presence in chronic lung disease of prematurity. *Thorax*. 2010;65(3):246–251. doi:10.1136/thx.2009.116061
28. Yang X, Zhang Y, Lin H, et al. Diagnostic value of the triple combination of serum heparin-binding protein, procalcitonin, and C-reactive protein in children with acute bacterial upper respiratory tract infection. *J Healthc Eng*. 2022;2022:1877960. doi:10.1155/2022/1877960
29. Eimonte M, Paulauskas H, Daniuseviciute L, et al. Residual effects of short-term whole-body cold-water immersion on the cytokine profile, white blood cell count, and blood markers of stress. *Int J Hyperther*. 2021;38(1):696–707. doi:10.1080/02656736.2021.1915504
30. Cooke J, Llor C, Hopstaken R, et al. Respiratory tract infections (RTIs) in primary care: narrative review of C reactive protein (CRP) point-of-care testing (POCT) and antibacterial use in patients who present with symptoms of RTI. *BMJ Open Respir Res*. 2020;7(1):e000624. doi:10.1136/bmjresp-2020-000624

International Journal of General Medicine

Publish your work in this journal

The International Journal of General Medicine is an international, peer-reviewed open-access journal that focuses on general and internal medicine, pathogenesis, epidemiology, diagnosis, monitoring and treatment protocols. The journal is characterized by the rapid reporting of reviews, original research and clinical studies across all disease areas. The manuscript management system is completely online and includes a very quick and fair peer-review system, which is all easy to use. Visit <http://www.dovepress.com/testimonials.php> to read real quotes from published authors.

Submit your manuscript here: <https://www.dovepress.com/international-journal-of-general-medicine-journal>

Dovepress
Taylor & Francis Group