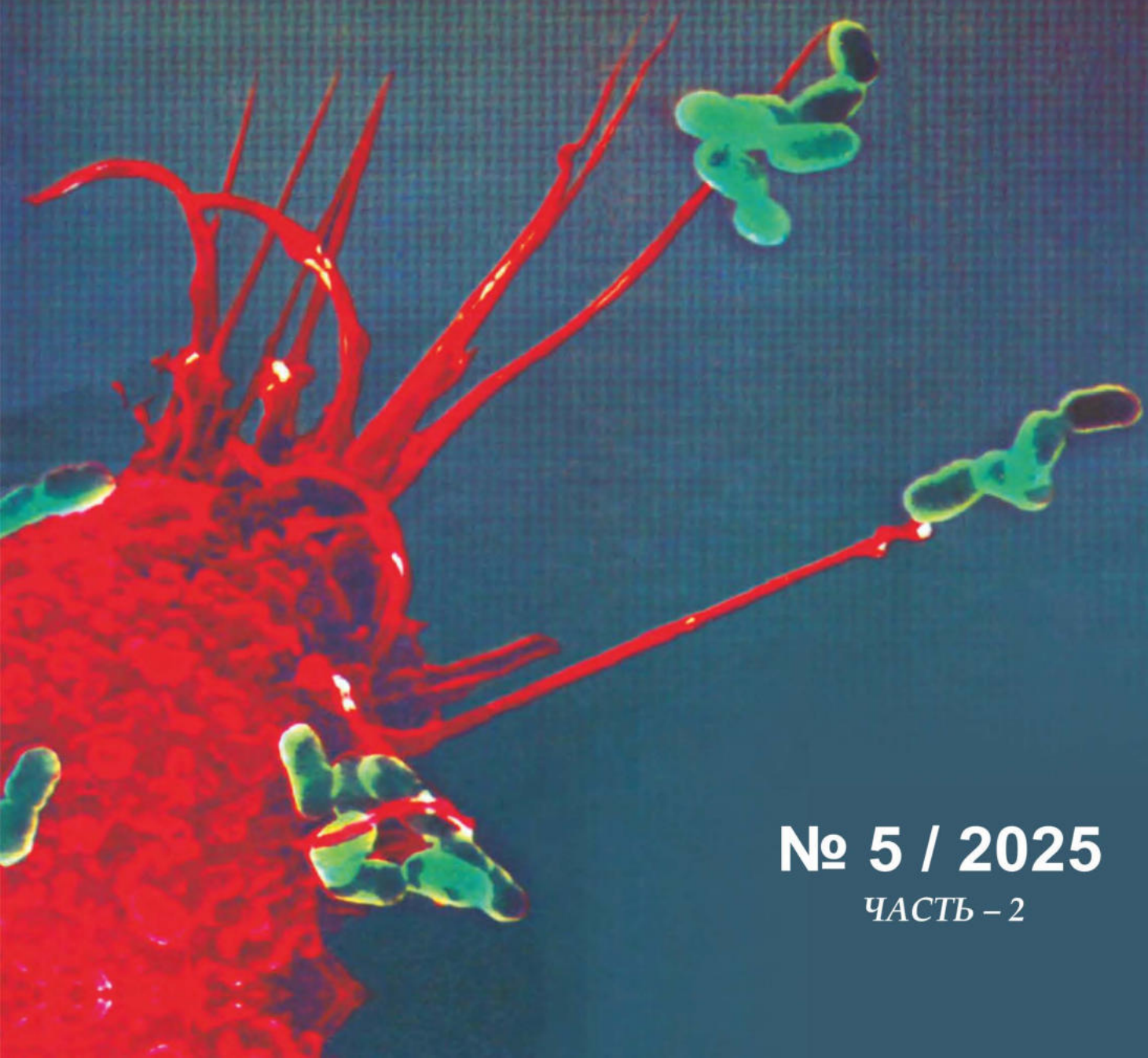


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MOLECULAR DIAGNOSTIC METHODS FOR ACUTE RESPIRATORY VIRAL INFECTIONS IN CHILDREN

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Relevance. Acute respiratory viral infections (ARVI) have remained the leading cause of morbidity among children for decades. According to the World Health Organization, more than one billion children experience various forms of ARVI annually, with a significant proportion occurring in children under 5 years of age [1]. In countries with temperate climates, each child has on average 6 to 10 episodes of infection per year, which imposes a substantial burden on healthcare systems and families [2].

Although most ARVI cases are mild to moderate, young children and patients with comorbidities are at increased risk of complicated disease, including pneumonia, bronchitis, sinusitis, and in some cases systemic infections [3,4]. Moreover, ARVI can exacerbate the course of chronic respiratory conditions such as bronchial asthma or cystic fibrosis [5].

In recent years, particular attention has been paid to viral–bacterial associations. Current evidence indicates that 25–30% of pediatric hospitalizations for severe ARVI are accompanied by bacterial co-infection [6]. The most common

bacterial pathogens include *Staphylococcus aureus*, *Streptococcus pneumoniae*, and *Haemophilus influenzae* [7]. Such associations significantly worsen the clinical course, prolong hospitalization, and increase the likelihood of antibiotic therapy.

Modern molecular genetic techniques, including PCR, have substantially expanded the possibilities of etiological diagnosis of respiratory infections [8]. However, the prognostic significance of pathogen detection remains an open question: to what extent a specific viral or bacterial agent influences the clinical course, frequency of complications, and duration of symptoms. In-depth investigation of this issue is of both theoretical and practical value for predicting outcomes, guiding rational antibiotic use, and informing preventive strategies [9].

Aim of the Study. To determine the prognostic significance of pathogen detection in children with ARVI and to assess the clinical course and laboratory features associated with different etiological variants of infection.

Materials and Methods. The study included 55 children aged 1 to 12 years who were hospitalized at Zangiota Specialized Infectious Diseases Hospital No. 1 and Tashkent City Children's Clinical Hospital No. 1 with a confirmed diagnosis of acute respiratory viral infection. The mean age was 5.1 ± 2.7 years; boys accounted for 29 (52.7%) and girls for 26 (47.3%). Diagnosis included clinical examination, assessment of body temperature, and evaluation of intoxication and catarrhal syndromes.

All patients underwent PCR testing for viral and bacterial pathogens of ARVI. Biological material consisted of oropharyngeal swabs collected using sterile probes under standard procedures. Samples were delivered to the PCR laboratory within 2–4 hours at $+2...+8$ °C. Nucleic acid extraction (RNA/DNA) was performed using commercial spin-column kits. For RNA viruses, reverse transcription was carried out to synthesize cDNA. Amplification was performed in real-time on a RotorGene 6000 thermal cycler using primers and probes specific for genomic sequences of Influenza A virus, Respiratory Syncytial Virus (RSV), Adenovirus, Bocavirus, as well as bacteria (*Staphylococcus aureus*, *Streptococcus oralis*). The PCR protocol included standard cycling: denaturation at 94–95 °C, annealing at 55–60 °C, and extension at 72 °C for 35–40 cycles. Results were assessed based on amplification curves with a threshold cycle (Ct) < 35. Positive and negative controls were included in each run.

Additional laboratory tests included complete blood count and C-reactive protein (CRP) levels. Data analysis was performed using IBM SPSS Statistics v25.0 (IBM Corp., Armonk, NY, USA).

Quantitative variables were expressed as mean \pm standard deviation (M \pm SD). Student's t-test was applied for comparison of quantitative variables between groups, and Pearson's χ^2 test for categorical variables. A p-value < 0.05 was considered statistically significant.

Results and Discussion. Etiological structure: viral infection was diagnosed in 34 children (61.8%), bacterial co-infections in 9 (16.4%), and no pathogen was detected in 21 (38.2%). The most frequent virus was Influenza A – 18 cases (32.7%), followed by RSV – 13 (23.6%), Adenovirus – 3 (5.5%), and Bocavirus – 3 (5.5%). Among bacterial pathogens, *Staphylococcus aureus* dominated with 5 cases (9.1%), while *Streptococcus oralis* was detected in 1 case (1.8%). Mixed viral–bacterial infections were found in 3 children (5.5%).

Clinical features: children with Influenza A more frequently presented with fever (37.5–38.0 °C), prominent catarrhal symptoms, and elevated CRP levels. RSV infection was associated with bronchial obstruction and prolonged disease duration (mean 7.5 ± 2.1 days vs. 5.2 ± 1.6 days for Influenza A, $p < 0.05$). Adenovirus infection was accompanied by conjunctivitis and more pronounced intoxication syndrome. Bocavirus infection had a milder course, with short-term subfebrile fever and weakly expressed symptoms.

Viral–bacterial associations were particularly notable. Children with Influenza A and *Staphylococcus aureus* co-infection showed the highest inflammatory marker levels: CRP – 45.6 ± 12.3 mg/L (vs. 12.4 ± 6.1 mg/L in isolated viral infection, $p < 0.01$). Fever duration was also longer – 6.8 ± 1.9 days compared to 3.5 ± 1.2 days ($p < 0.05$). These

findings align with previous reports highlighting the aggravating role of *Staphylococcus aureus* in viral infections [10,11].

Table 1.

Etiological Structure and Clinical Features of ARVI in Children (n=55)

Pathogen	Cases (n)	%	Key Clinical Features
Influenza A	18	32.7%	Fever, catarrhal syndrome
RSV	13	23.6%	Bronchial obstruction, prolonged course
Adenovirus	3	5.5%	Intoxication, conjunctivitis
Bocavirus	3	5.5%	Mild, subclinical course
<i>Staphylococcus aureus</i>	5	9.1%	Exacerbated inflammation, complications
<i>Streptococcus oralis</i>	1	1.8%	Localized complications
Mixed infections	3	5.5%	Prolonged fever, severe course
None detected	21	38.2%	Mild/moderate course

Our findings are consistent with international studies. Shi et al. [12] reported RSV as the leading cause of hospitalization among young children. Influenza A is associated with pronounced catarrhal symptoms and frequent pneumonia [13]. Adenovirus infection is described as highly intoxicating, often with conjunctivitis and lymphadenopathy [14]. Bocavirus is typically mild but can aggravate the course when co-occurring with other viruses [15].

Particular emphasis is placed on viral-bacterial co-infections. Studies by Caballero et al. [16] and Martin et al. [17] showed that combinations with *Staphylococcus aureus* and *Streptococcus pneumoniae* lead to the most severe outcomes. Our data support these conclusions: children with such associations had higher complication rates, longer hospital stays, and more severe courses. Thus, pathogen type directly influences

the prognosis of ARVI in children. In the context of increasing antimicrobial resistance, early detection of mixed infections and risk stratification become critically important.

Conclusion. This study demonstrated that the prognosis of ARVI in children largely depends on the etiological agent. RSV infection is linked with bronchial obstruction and prolonged disease. Influenza A is accompanied by pronounced catarrhal syndrome and elevated inflammatory markers. Adenovirus induces more severe intoxication, while Bocavirus typically follows a mild course. The most unfavorable outcomes were observed in viral-bacterial associations, particularly Influenza A with *Staphylococcus aureus*.

These results confirm the necessity of routine PCR diagnostics in children with ARVI to determine etiology and predict disease progression. This approach

allows for identification of high-risk groups, rational use of antibiotics, and reduction of adverse outcomes.

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РЕЗЮМЕ

МЕТОДЫ МОЛЕКУЛЯРНОЙ
ДИАГНОСТИКИ ОСТРЫХ
РЕСПИРАТОРНЫХ ВИРУСНЫХ
ИНФЕКЦИЙ У ДЕТЕЙ

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Ключевые слова: острые респираторные инфекции, дети, ПЦР, бактериология, коинфекция, диагностика, антимикробная резистентность.

В исследование включены 55 пациентов в возрасте от 1 до 12 лет, у которых с помощью ПЦР определялись вирусные и бактериальные патогены. Этиологическое распределение было следующим: Influenza A – 32,7%, RSV – 23,6%, аденовирус – 5,5%, боксавирус – 5,5%, Staphylococcus aureus – 9,1%, Streptococcus oralis – 1,8%, смешанные инфекции – 5,5%, отсутствие патогена – 38,2%.

Заключение: исход ОРВИ у детей во многом определяется видом возбудителя, а использование ПЦР позволяет своевременно выявлять группы риска и корректировать тактику лечения.

REZUME

BOLALARDA O'TKIR RESPIRATOR
VIRUSLI INFEKSIYALARNI
MOLEKULAR DIAGNOSTIKA
USULLARI

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Khudaykulova Gulnara Karimovna,
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Kalit so'zlar: o'tkir respirator infeksiyalar, bolalar, PZR, bakteriologiya, qo'shaloq infeksiya, diagnostika, antimikrob rezistentlik

PZR usuli yordamida virusli va bakterial patogenlar aniqlangan 1–12 yoshdagi 55 nafar bemor o'rganildi. Etiologik tuzilma quyidagicha bo'ldi: A grippi – 32,7%, RSV – 23,6%, adenovirus – 5,5%, bocavirus – 5,5%, Staphylococcus aureus – 9,1%, Streptococcus oralis – 1,8%, aralash infeksiyalar – 5,5%, qo'zg'atuvchi aniqlanmagan holatlar – 38,2%.

Xulosa: O'RVVI prognozi bolalarda asosan qo'zg'atuvchiga bog'liq bo'lib, PZR diagnostikasi xavf guruhlarini aniqlash va davolashni optimallashtirishda muhim ahamiyatga ega.

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