viruses

MDPI

*Article*

# Reported Clinical Spectrum of Pediatric Human Adenovirus Type 14 Infection Across Case-Ascertainment Settings: A Case-Level Synthesis and Genomic Characterization of a Severe-Case Isolate

**Jiangyu Cai [1,†], Na Zang [1,†], Ying Lu [1], Zhongying Yang [1], Fuping Yang [1], Ying Yang [1], Hui Qu [1], Yushun Wan [1,2], Yueqiang Fu [3,*] and Enmei Liu [1,*]**

[1] Department of Respiratory Medicine, Children's Hospital of Chongqing Medical University, National Clinical Research Center for Children and Adolescents' Health and Diseases, Ministry of Education Key Laboratory of Child Development and Disorders, International Science and Technology Cooperation Base of Child Development and Critical Disorders, Chongqing Key Laboratory of Child Rare Diseases in Infection and Immunity, Chongqing 400014, China
[2] College of Basic Medicine, Chongqing Medical University, Chongqing 400016, China
[3] Department of Critical Care Medicine, Children's Hospital of Chongqing Medical University, Chongqing 400014, China
* Correspondence: 480887@hospital.cqmu.edu.cn (Y.F.); emliu186@126.com (E.L.)
† These authors contributed equally to this work.

## Abstract

Human adenovirus type 14 (HAdV-14) is an uncommon respiratory adenovirus associated with outbreaks and severe disease, but its clinical spectrum in children remains poorly defined. We conducted a structured case-level synthesis of pediatric HAdV-14 infections identified in the literature and integrated two locally identified cases to characterize clinical presentation, disease severity, and host factors associated with adverse outcomes. The critically ill local case underwent longitudinal clinical profiling, virus isolation, whole-genome sequencing, and phylogenetic and comparative genomic analyses. Among the 168 pediatric cases identified, all 111 outbreak-associated cases were non-severe, whereas among the 43 classifiable sporadic cases, 34 were non-severe, 7 were severe, and 2 were fatal. In an exploratory analysis, adverse premorbid conditions were more frequently reported in severe/fatal than in non-severe sporadic cases (55.6% vs. 2.9%; unadjusted OR, 41.25; 95% CI, 3.80–447.78; $p = 0.001$). The local cases ranged in severity from self-limited bronchitis to critical pneumonia with long-term sequelae. The severe-case isolate belonged to HAdV-14p1-II and was highly similar to previously reported HAdV-14p1 strains. Comparative analysis of seven HAdV-14 whole-genome sequence records with available pediatric clinical information showed that severe/fatal isolates occurred in both HAdV-14p1 clades (I and II), with no evident consensus-level genomic variant pattern distinguishing severe/fatal from non-severe infections. These findings indicate that the reported severity of pediatric HAdV-14 infection varies across case-ascertainment settings. The observed differences in reported premorbid conditions and the available genomic findings suggest that host vulnerability and ascertainment context should be considered when interpreting disease severity, rather than attributing severe outcomes to HAdV-14 type or lineage alone.

Academic Editor: Theo J. Moraes

Received: 14 October 2026
Revised: 22 November 2026
Accepted: 25 November 2026
Published: 29 November 2026

**Copyright:** © 2026 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license.

**Keywords:** human adenovirus type 14; HAdV-14; pediatric infection; severe disease; clinical spectrum; genomic epidemiology; host vulnerability

## 1. Introduction

Human adenoviruses (HAdVs) are important causes of respiratory infection in children, with clinical manifestations and disease severity varying substantially across viral types [1]. Human adenovirus type 14 (HAdV-14), a historically uncommon member of species *Human mastadenovirus B*, has received particular attention because it can cause severe and fatal respiratory disease. First identified during an outbreak among military recruits in the Netherlands in 1955, HAdV-14 was infrequently detected for several decades before a distinct genome type, HAdV-14p1, emerged in North America in the early 2000s [2]. During the 2006–2007 US outbreaks, 38% of 140 confirmed cases were hospitalized, 17% required intensive care, and 5% died [3]. Subsequent reports of community outbreaks also documented substantial morbidity and mortality, contributing to the perception of HAdV-14 as a highly pathogenic respiratory adenovirus [4].

However, the severe outcomes highlighted in early reports may not represent the full clinical spectrum of HAdV-14 infection. Among otherwise healthy military trainees with pneumonia, HAdV-14 infection was not associated with greater pneumonia severity, intensive care admission, longer hospitalization, or 30-day mortality compared with pneumonia not associated with HAdV-14 [5]. Moreover, HAdV-14 outbreaks have varied markedly in severity despite highly similar viral genomes, and whole-genome comparison of isolates from mild respiratory illness and fatal pneumonia identified no clear viral genetic determinant of disease severity [6]. These apparently divergent observations raise an important question: to what extent does the severe disease historically associated with HAdV-14 represent the broader clinical expression of infection, and what clinical or epidemiological factors may be associated with the observed variation in disease severity?

The clinical spectrum of HAdV-14 infection in children remains poorly characterized. HAdV-14 is an uncommon type among pediatric adenovirus infections, making it difficult to establish sufficiently large type-specific pediatric cohorts. This challenge is compounded by limited adenovirus typing in routine clinical practice, as typing generally does not influence clinical management and is more frequently performed in the context of severe infections or outbreaks [7,8]. Most detailed epidemiological evidence comes from military populations, community outbreaks, or mixed-age cohorts. Consequently, evidence of pediatric HAdV-14 remains fragmented, and observations in predominantly healthy young adults cannot necessarily be extrapolated to children.

To address this gap, we conducted a structured case-level synthesis integrating previously reported pediatric HAdV-14 infections with two locally identified cases; the critically ill local case also underwent longitudinal clinical profiling and genomic characterization of the corresponding isolate. Using published pediatric evidence alongside local clinical and virological observations, we aimed to delineate the reported clinical spectrum and severity profile of pediatric HAdV-14 infection, explore potential factors associated with severe outcomes, and provide an overview of the available pediatric evidence to inform future studies.

## 2. Materials and Methods

### *2.1. Literature Search and Case Extraction*

A structured literature search was conducted in PubMed, Web of Science, Embase, CNKI, and Wanfang from database inception to February 2026 to identify reports of

pediatric HAdV-14 infections. The search terms included combinations of "human adenovirus 14", "HAdV-14", "adenovirus type 14", "adenovirus 14", "Adv14", "ADV-14", "adenovirus serotype 14" and corresponding Chinese terms. Reference lists of relevant articles were also manually screened for additional eligible reports. Full search strategies are provided in the Supplementary Methods, and the study-selection process is shown in Figure S1.

Cases were eligible if patients were younger than 18 years with HAdV-14 identified in respiratory specimens and had extractable demographic, clinical, outcome, or epidemiological information. Reports without confirmed pediatric HAdV-14 infection, reviews without original case data, and duplicate cases were excluded. For overlapping reports, the report providing the most complete information was retained.

Available demographic, clinical, outcome, and epidemiological data were extracted for each eligible case. Based on the epidemiological context described in the original publication, cases explicitly reported as part of an HAdV-14-associated outbreak or an epidemiologically linked cluster were classified as outbreak-associated cases; cases reported outside such outbreaks or clusters, or without a reported epidemiological link to an outbreak or cluster, were classified as sporadic cases. This classification was informed by the Chinese Technical Guidelines for Prevention and Control of Human Adenovirus Respiratory Infection (2019 Edition) [9], which specify reporting thresholds for clustered respiratory infections (≥3 cases in the same class within one day or ≥5 cases within three days) and adenovirus outbreaks (≥30 clinically diagnosed cases or ≥5 laboratory-confirmed cases within one week in the same collective setting or village), with additional criteria for laboratory-confirmed critically ill or fatal cases. Methodological quality and risk of bias were assessed independently by two reviewers using the appropriate Joanna Briggs Institute critical appraisal tools according to study design. Details of the assessment procedure are provided in the Supplementary Methods and Tables S1–S5.

### *2.2. Case Definitions and Statistical Analysis*

The severity classification was informed by the 2024 revised Chinese Guidelines for the Management of Community-Acquired Pneumonia in Children [10], with operational criteria adapted to account for the heterogeneous clinical presentations and incomplete reporting of the included cases. Cases were classified as severe if they met at least one of the following criteria: (1) an explicit diagnosis of severe pneumonia, acute respiratory distress syndrome (ARDS), respiratory failure, or sufficient clinical evidence supporting severe respiratory disease; (2) admission to an ICU/PICU; (3) requirement for invasive mechanical ventilation (IMV), acute non-invasive ventilation (NIV) or advanced respiratory support such as extracorporeal membrane oxygenation (ECMO) for acute respiratory dysfunction; (4) development of severe pulmonary or extrapulmonary complications associated with the acute disease course. Fatal cases were classified separately. Cases were classified as non-severe when the original reports explicitly described mild illness, provided sufficient clinical information supporting a non-severe classification, or clearly documented the absence of severe disease or related adverse outcomes.

Statistical analysis and visualization were performed using R version 4.4.3 and Python version 3.13.2. Continuous variables are presented as medians with interquartile ranges and categorical variables as frequencies and percentages. Exploratory comparisons between non-severe and severe/fatal sporadic cases were performed using the Wilcoxon rank-sum test for continuous variables and Fisher's exact test for categorical variables, where applicable. No formal risk modelling was performed because of the small number and heterogeneous reporting of available cases.

### *2.3. Local Cases and Clinical Data Collection*

Two children with laboratory-confirmed HAdV-14 infection identified at Children's Hospital of Chongqing Medical University were included as complementary local cases. For Case 1, HAdV-14 was identified by PCR amplification and Sanger sequencing of the *penton base*, *hexon*, and *fiber* genes, followed by sequence comparison with reference sequences using the NCBI BLASTn web tool (National Center for Biotechnology Information, Bethesda, MD, USA; https://blast.ncbi.nlm.nih.gov/). For Case 2, HAdV-14 was identified by whole-genome sequencing and subsequent sequence comparison and phylogenetic analyses (as described below). Demographic characteristics, clinical manifestations, laboratory and radiological findings, treatments, complications, sequelae, and outcomes were retrospectively extracted from electronic medical records. The critically ill child also underwent longitudinal clinical profiling, and a clinical virus isolate was obtained for genomic characterization. The two local cases were incorporated into the case-level dataset and classified as sporadic for the descriptive and exploratory analyses.

This study was approved by the Ethics Committee of Children's Hospital of Chongqing Medical University (approval number: 2024 Ethics Review [Research] No. [217-1]). Written informed consent was obtained from the parents or legal guardians for the use of clinical data and residual clinical specimens.

### *2.4. Virus Isolation and Genome Sequencing*

Virus isolation was performed using the nasopharyngeal aspirate sample from the local severe case. The sample was collected from Case 2 on hospital day 2 and then inoculated onto A549 cells (ATCC, Manassas, VA, USA) and passaged twice. Infected cell cultures were subjected to three freeze–thaw cycles, followed by viral genomic DNA extraction. Because no viable specimens were available from the earlier case, virus isolation and whole-genome sequencing were performed only for the recently identified severe case.

Whole-genome sequencing was performed on a DNBSEQ-G400 platform (Wuhan MGI Tech Co., Ltd., Wuhan, China) at Beijing Genomics Institute (BGI, Shenzhen, China). Genomic DNA was randomly sheared to construct paired-end libraries with insert sizes of approximately 300–400 bp. After quality filtering, clean reads were used for genome assembly and downstream analyses. Further details are provided in the Supplementary Methods.

### *2.5. Genome Assembly, Annotation and Completeness Assessment*

Sequencing reads underwent quality assessment and were filtered to remove host-derived reads. Non-human read pairs were assembled de novo, and the dominant adenovirus-B-like contig was selected for downstream annotation and genomic analysis. Genome completeness was assessed based on assembly length, internal ambiguous bases, coding-region integrity, terminal regions, and sequencing depth, including assessment of the *penton base*, *hexon*, and *fiber* coding regions. The resulting assembly was designated a coding-complete HAdV-14 genome with unresolved termini. Detailed bioinformatic procedures are provided in the Supplementary Methods.

### *2.6. Phylogenetic and Comparative Genomic Analyses*

CQ202503 was compared with publicly available HAdV-14 whole-genome sequences retrieved from GenBank. Maximum-likelihood phylogenetic analyses of the whole genome and the *penton base*, *hexon*, and *fiber* genes were performed to determine its molecular placement and type assignment. The reference sequences used for phylogenetic analysis and their GenBank accession numbers are listed in Table S6.

Whole-genome pairwise sequence identity was calculated between CQ202503 and selected representative HAdV-14 genomes, including the historical prototype strain de

Wit, the representative international HAdV-14p1 strain USA OR2971 2007, and previously reported Chinese HAdV-14p1 strains (Beijing 2010, Beijing 2012, GZ01 2010, GZ02 2010, and Gansu 2013). Coding-region nucleotide and amino acid differences were further characterized using the CQ202503 coordinate system.

For the clinical-genomic analysis, seven HAdV-14 whole-genome sequence records with available pediatric clinical information were selected, including USA/AK2314/2005 (MG642743.1), Ireland/1782/2010 (MG905100.1), 5058S (MG905101.1), BJ430 (JN032132.1), GZ01 (JQ824845.1), Kobe-240057 (LC830688.1), and the locally sequenced isolate CQ202503 (PZ397459). Clinical severity was defined as previously described. Pairwise whole-genome single-nucleotide polymorphism (SNP) differences among the seven clinical-information-linked isolates were calculated using aligned nucleotide positions with unambiguous bases (A, C, G, and T) in both sequences. To characterize variant-sharing patterns, the seven isolates were compared with the historical HAdV-14 prototype genome AY803294.1. Single-nucleotide variants and insertions/deletions were identified at callable alignment positions and mapped to prototype-genome coordinates. Coding variants were annotated using the GenBank coding sequence features. Variant-sharing patterns were classified as shared across clinical phenotype categories, isolate-specific, shared by all three severe/fatal isolates but absent from both non-severe isolates, or shared by both non-severe isolates but absent from all severe/fatal isolates. These analyses were descriptive; no formal statistical testing was performed.

Detailed procedures for sequence alignment, phylogenetic reconstruction, pairwise identity calculation, and coding-sequence comparison are provided in the Supplementary Methods.

## 3. Results

### *3.1. Characteristics and Distribution of Available Pediatric HAdV-14 Cases*

The literature search identified 18 eligible publications reporting 166 pediatric HAdV-14 cases [2,3,11–26]. After inclusion of the two local cases, the case-level analysis included 168 cases. Of these, 57 were sporadic and 111 were outbreak-associated, as described in the original reports (Tables S7 and S8). Most reported cases originated from China (151/168, 89.9%), followed by the United States (9/168, 5.4%), Ireland (6/168, 3.6%), Canada (1/168, 0.6%) and Japan (1/168, 0.6%) (Figure 1A). Within China, cases were reported from multiple regions, with the largest numbers from Jiangsu (49/151, 32.5%), Gansu (43/151, 28.5%), and Liaoning (24/151, 15.9%) (Figure 1B).

Reports of pediatric HAdV-14 infections spanned several decades. Sporadic cases were concentrated between 2001 and 2015, accounting for 50 of 57 (87.7%) sporadic cases. Relatively few sporadic cases were reported after 2016, including one case in early 2019 and three additional cases during 2024–2025. All outbreak-associated cases included in this study were reported between 2011 and 2015 (Figure 1C). Among cases with available month-of-infection data, pediatric HAdV-14 infections were reported throughout the year, with more cases reported in spring and summer; the proportion classified as severe among sporadic cases also varied across seasons (Figure 1D). Males accounted for 62.2% of sporadic cases and 55.9% of outbreak-associated cases with available sex information (Figure 1E). Outbreak-associated cases predominantly involved school-aged children (Table S7), whereas sporadic cases spanned a broader age range and included a substantial proportion of children younger than 2 years (Figure 1F). These temporal and seasonal patterns are descriptive and were not subjected to formal trend analysis.

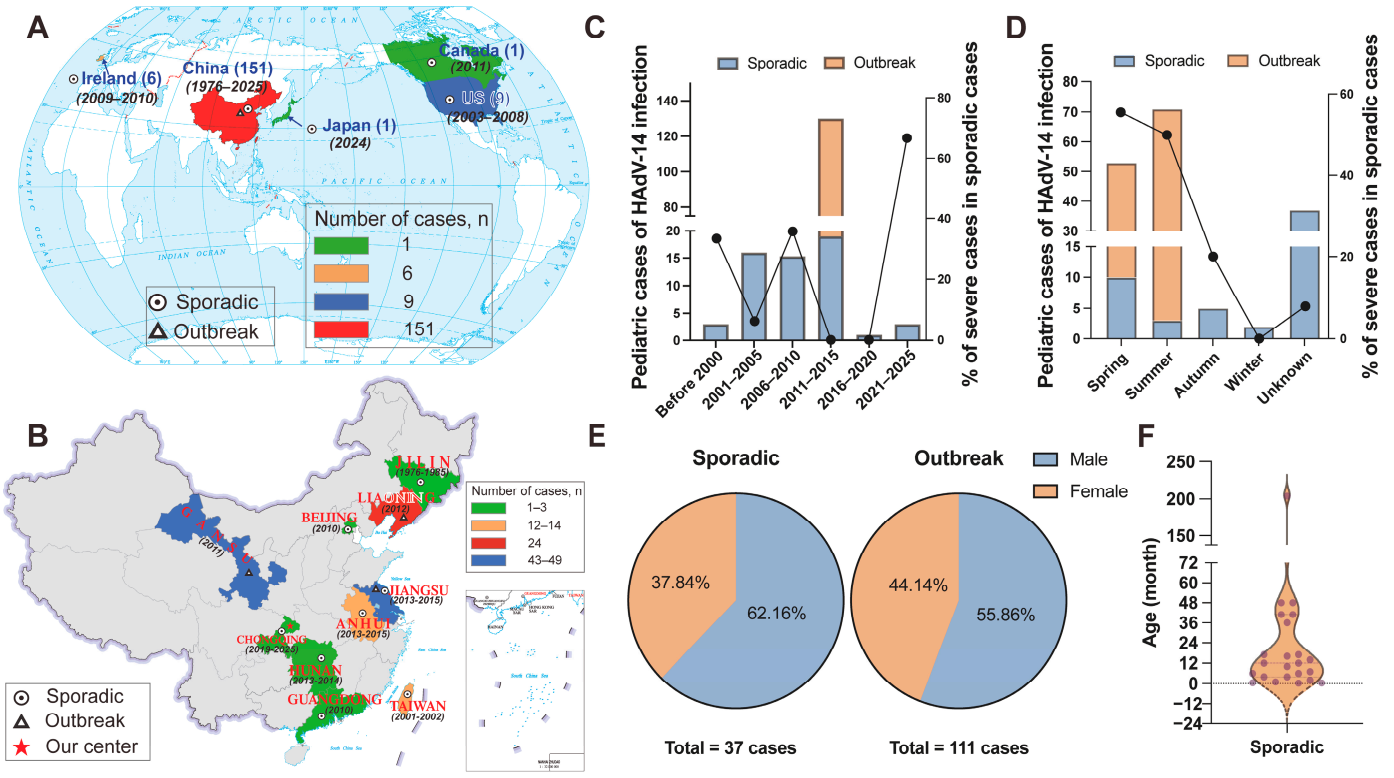



**Figure 1.** Geographic, temporal and demographic distribution of reported pediatric HAdV-14 cases. (**A,B**) Geographic distribution of reported pediatric HAdV-14 cases worldwide and in China. Circles indicate sporadic cases, and triangles indicate outbreak-associated cases. (**C**) Temporal distribution of reported pediatric HAdV-14 cases by year or reporting period. Connected points indicate the proportion of severe cases among sporadic cases in each period. (**D**) Seasonal distribution of reported pediatric HAdV-14 cases with available infection-month information. Seasons were defined according to Northern Hemisphere meteorological seasons. Points connected by a line indicate the proportion of severe cases among sporadic cases in each season. (**E**) Sex distribution of sporadic and outbreak-associated pediatric HAdV-14 cases. (**F**) Age distribution of reported sporadic pediatric HAdV-14 cases.

### *3.2. Reported Clinical Spectrum and Disease Severity Differed by Case-Ascertainment Setting*

The reported clinical spectrum differed markedly between outbreak-associated and sporadic pediatric HAdV-14 cases. All 111 outbreak-associated cases included in the analysis were non-severe, with reported diagnoses predominantly comprising upper respiratory tract infections or febrile respiratory illness; no severe or fatal cases were reported (Figure 2A,B; Table S7). Common manifestations included fever, cough, sore throat, rhinorrhea, and headache. In contrast, sporadic cases showed a broader clinical spectrum, ranging from non-severe respiratory illness to severe pneumonia, ARDS, ECMO-supported critical illness, neurological involvement, and death (Figure 2A,B; Table S8). Among sporadic cases with detailed clinical descriptions, wheezing, dyspnea or respiratory distress, hypoxemia or cyanosis, and gastrointestinal or neurological manifestations were also reported [16–18,20,21], whereas co-detection of other pathogens was infrequently documented (Tables S7 and S8).

Among the 43 sporadic cases for which disease severity could be classified, 34 were non-severe, 7 were severe, and 2 were fatal. Co-detection status was unknown in 28/34 (82.4%) non-severe cases and 4/9 (44.4%) severe/fatal cases. Among cases with documented testing results, other pathogen co-detection was reported in 0/6 non-severe cases and 1/5 severe/fatal cases (Table 1). No complications or long-term sequelae were reported among the non-severe cases. In contrast, complications were reported in 4 of 9 (44.4%)

severe/fatal cases, including bronchiectasis, pleural effusion, lung abscess, encephalitis or toxic encephalopathy, and HIBD (Figure 2C). Five of nine (55.6%) severe/fatal cases survived with reported sequelae, including BO and neurological or developmental sequelae, and the two fatal cases occurred in female infants younger than 1 month (Figure 2D).

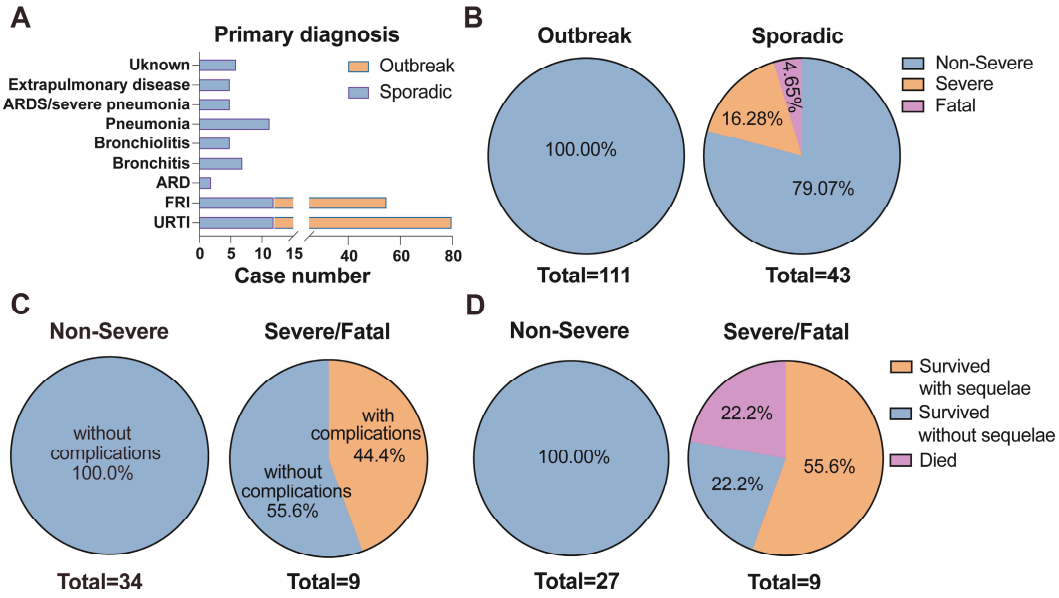



**Figure 2.** Clinical spectrum, disease severity, complications, and outcomes of reported pediatric HAdV-14 infections. (**A**) Distribution of primary clinical diagnoses among outbreak-associated and sporadic pediatric HAdV-14 cases. Diagnostic categories were not mutually exclusive. (**B**) Distribution of disease severity among outbreak-associated and sporadic cases. (**C**) Distribution of reported complications among non-severe and severe/fatal sporadic cases. (**D**) Reported outcomes among non-severe and severe/fatal sporadic cases. Abbreviations: ARD, acute respiratory disease; ARDS, acute respiratory distress syndrome; FRI, febrile respiratory illness; URTI, upper respiratory tract infection.

**Table 1.** Exploratory comparison of reported sporadic pediatric HAdV-14 cases by disease severity.

| Variable | Category | Non-Severe (*n*/*N*, %) * | Severe/Fatal (*n*/*N*, %) * | *p* Value | Effect Size (95% CI) |
|---|---|---|---|---|---|
| Age, months, median (Q1, Q3) | / | 14.5 (6.0, 46.3) | 14.0 (0.5, 17.0) | 0.360 | rank-biserial correlation = 0.238 |
| Sex, *n* (%) | Male<br>Female | 16/26 (61.5)<br>10/26 (38.5) | 5/9 (55.6)<br>4/9 (44.4) | 1.000 | OR = 0.78 (0.17–3.62) |
| Season, *n* (%) | Spring<br>Summer<br>Autumn<br>Winter | 4/11 (36.4)<br>1/11 (9.1)<br>4/11 (36.4)<br>2/11 (18.2) | 5/7 (71.4)<br>1/7 (14.3)<br>1/7 (14.3)<br>0/7 (0.0) | 0.506 | Cramér's *V* = 0.420 |
| Co-detection, *n*/*N* (%) | / | 0/6 (0.0) | 1/5 (20.0) | 0.455 | Risk difference = 20.0 pp (−22.3 to 62.4) |
| Adverse premorbid condition, *n* (%) | / | 1/34 (2.9) | 5/9 (55.6) | 0.001 | OR = 41.25 (3.80–447.78) |

* Comparisons were exploratory and based on extractable information from reported sporadic pediatric HAdV-14 cases. Denominators vary across variables because of missing or incompletely reported data. *p* values were calculated using Wilcoxon rank-sum tests for continuous variables and

Fisher's exact tests for categorical variables, where applicable. No formal risk modelling was attempted because of the sparse and heterogeneous nature of the data. OR, odds ratio; CI, confidence interval; pp, percentage points.

### *3.3. Adverse Premorbid Conditions Were Associated with Severe/Fatal Disease in Sporadic Cases*

Exploratory comparisons between non-severe and severe/fatal sporadic cases did not identify statistically significant differences in age, sex, season or co-detection (Table 1). Adverse premorbid conditions, however, were reported more frequently among severe/fatal than non-severe cases (5/9 [55.6%] vs. 1/34 [2.9%]; unadjusted OR = 41.25; 95% CI, 3.80–447.78; $p$ = 0.001). Reported premorbid conditions among severe/fatal cases included prematurity, recurrent hospitalization for lower respiratory tract infection, natural killer cell deficiency, developmental delay, and suspected pathogenic chromosomal copy-number variations (Tables 1 and S8). Given the limited number of cases and heterogeneous source data, this association was considered exploratory rather than evidence of an independent risk factor. Although co-detection did not differ significantly between severe/fatal and non-severe cases, the available data remain limited (Table 2), and secondary infections may occur during the clinical course. Therefore, severe clinical manifestations and adverse outcomes cannot be attributed to HAdV-14 infection alone with certainty.

**Table 2.** Clinical and microbiological characteristics of two local pediatric HAdV-14 cases and eight additional severe/fatal cases reported in the literature.

| Cases | Age (Months) | Severity | Diagnosis/Key Clinical Findings | Other Pathogens Tested | Co-Detection | Complications | Outcome |
|---|---|---|---|---|---|---|---|
| †1 | 41 | Non-severe | Bronchitis; Fever, cough, nasal obstruction | bacterial cultures; RSV, IV-A, IV-B, PIV 1–3 antigen tests | Negative | NR | Survived without sequelae |
| †‡2 | 14 | Severe | Severe pneumonia; ARDS; necrotizing pneumonia; Sepsis | bacterial cultures; RSV, IV-A, IV-B, PIV 1–3 antigen tests; mNGS | HRV, *H. influenzae*, *S. pneumoniae*, *S. maltophilia*, *A. xylosoxidans* | Pleural effusion, Thrombosis, HIBD * | Survived with brain atrophy, GDD, bronchiolitis obliterans |
| 3 [2,3] | 10 | Severe | Pneumonia | NR | Unknown | NR | Survived with pulmonary sequelae |
| 4 [2,3] | 0.4 | Fatal | Adenoviral bronchiolitis; ARDS; dehydration, poor feeding | NR | Unknown | NR | Died |
| 5 [16] | 17 | Severe | Pneumonia; acute tracheobronchitis; $SpO_2$ 80%; multilobe infiltrates. | IV and RSV antigen tests; viral and bacterial cultures | Negative | right upper lobe bronchiectasis | Survived with chronic bronchiolitis obliterans |
| 6 [16] | 36 | Severe | ARDS; fever; $SpO_2$ 70%; respiratory distress | blood, urine, and ETA bacterial cultures; respiratory viral DFA | Bacterial cultures negative; respiratory viral DFA unrevealing | bronchiectasis; right upper-lobe abscess | Survived; required prolonged ventilation, tracheostomy, and gastrostomy feeding |
| 7 [17,18] | 0.3 | Fatal | Pneumonia; hypotonic, abnormal liver function test results | IFA for IV-A, IV-B, PIV 1–3, and RSV | Negative | NR | Died |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 8 [17,18] | 0.5 | Severe | ARDS; plasma adenoviral load of 8.83 $\log_{10}$ viral genomes/mL | IFA for IV-A, IV-B, PIV 1–3, and RSV | Negative | NR | Survived |
| 9 [20] | 84 | Severe | Acute encephalopathy; fever, cough, and seizures; $SpO_2$ 94%; GCS 9; no pneumonia | Nasopharyngeal multiplex PCR (other targets NR) | Unknown | NR | Survived with upper limb weakness at discharge, no neurologic problems at follow-up 1 month after diagnosis |
| 10 [21] | 16 | Severe | Pneumonia; fever, cough, wheezing; cyanosis; respiratory failure | NR | Unknown | Heart failure, respiratory failure, toxic encephalopathy | Survived |

Notes: Co-detection results are presented according to the pathogen testing information available in the original reports or clinical records. Negative results apply only to the specified tests and do not exclude pathogens that were not assessed. Unknown indicates that the detection status of additional pathogens could not be determined from the available information. † Cases identified at our hospital. ‡ CQ202503, the HAdV-14 isolate characterized by whole-genome sequencing in this study. * Neurologic injury was clinically attributed to hypoxic–ischemic injury after cardiopulmonary resuscitation rather than direct viral CNS infection. Abbreviations: NR, not reported; HRV, human rhinovirus; ARDS, acute respiratory distress syndrome; HIBD, hypoxic–ischemic brain damage; ECMO, extracorporeal membrane oxygenation; IMV, invasive mechanical ventilation; CPAP, continuous positive airway pressure; IVIG, intravenous immunoglobulin; GDD, global developmental delay; RSV, respiratory syncytial virus; IV, influenza virus; PIV, parainfluenza virus; $SpO_2$, peripheral oxygen saturation; ETA, endotracheal aspirate; DFA, direct fluorescent antibody assay; IFA, immunofluorescence assay; mNGS, metagenomic next-generation sequencing; GCS, pediatric Glasgow Coma Scale.

### *3.4. Local Pediatric HAdV-14 Cases as Complementary Clinical Observations*

Two children with laboratory-confirmed HAdV-14 infection were identified at Children's Hospital of Chongqing Medical University and showed markedly different clinical presentations, disease severity, treatment requirements, and outcomes (Table 2).

Case 1 was a 41-month-old girl with bronchitis who presented with nasal obstruction, cough, and fever. Laboratory abnormalities were limited to an increased neutrophil percentage, a reduced lymphocyte percentage, and mildly elevated procalcitonin (Table 3), and chest imaging showed no evidence of substantial lower respiratory tract involvement (Figure 3A,B). She improved with supportive care and was discharged after 3 days. Case 2 was a 14-month-old boy admitted with cough, fever, dyspnea, and wheezing who developed a prolonged critical illness requiring 67 days of hospitalization. He was diagnosed with severe pneumonia, ARDS, necrotizing pneumonia, sepsis, and hypoxic–ischemic brain damage (HIBD) (Table 2). Admission laboratory findings showed thrombocytopenia, anemia, lymphopenia, increased neutrophil percentage, elevated inflammatory and coagulation markers, hypoalbuminemia, increased aspartate aminotransferase (AST) and lactate dehydrogenase (LDH) levels (Table 3). Chest radiography on admission showed diffuse bilateral pulmonary opacities consistent with severe pneumonia (Figure 3C).

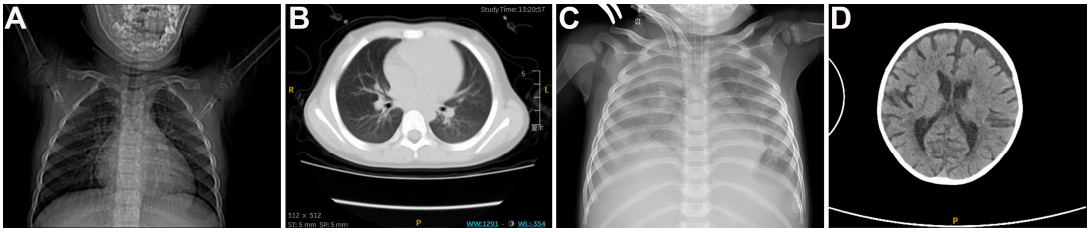



**Figure 3.** Radiological findings of two pediatric HAdV-14 cases identified in our hospital. (**A**,**B**) Chest radiograph and chest computed tomography image of Case 1 on the day of admission, showing no obvious evidence of severe lower respiratory tract involvement; (**C**) chest radiograph of Case 2 on the day of admission, showing diffuse bilateral pulmonary opacities consistent with severe pneumonia; (**D**) brain computed tomography of Case 2 performed 8 days after admission, showing cerebral atrophy and subdural effusion.

**Table 3.** Admission laboratory findings of two pediatric HAdV-14 cases in our hospital.

| Laboratory Parameter | Case 1 | Case 2 |
|---|---|---|
| **Routine blood indicators (units, reference range)** | | |
| White blood cell count (×10⁹/L, 5.1–14.1) | 10.75 | 9.01 |
| Platelet count (×10⁹/L, 100–524) | 239 | 77 |
| Red blood cell count (×10¹²/L, 4.10–5.50) | 4.48 | 3.36 |
| Hemoglobin (g/L, 107–141) | 126 | 101 |
| Absolute lymphocytes (×10⁹/L, 2.40–8.70) | 3.01 | 1.54 |
| Absolute neutrophils (×10⁹/L, 0.80–5.80) | 7.2 | 7.28 |
| Absolute monocytes (×10⁹/L, 0.18–1.13) | 0.43 | 0.17 |
| Percentage of lymphocytes (%, 33.0–77.0) | 28 | 17.1 |
| Percentage of neutrophils (%, 13.0–55.0) | 67 | 80.8 |
| Percentage of monocytes (%, 2.0–13.0) | 4 | 1.9 |
| **Inflammatory indicators (units, reference range)** | | |
| C-reactive protein (CRP, mg/L, <8) | <8 | 44.83 |
| Procalcitonin (PCT, ng/mL, <0.05) | 0.113 | 2.56 |
| D-dimer (DD, mg/L, <0.91) | / | 3.6 |
| **Blood biochemistry (units, reference range)** | | |
| Total protein (g/L, 56.2–72.9) | 68.2 | 54.4 |
| Albumin (g/L, 38.3–53.2) | 41.8 | 30.3 |
| Alanine aminotransferase (ALT, U/L, 11.9–44.9) | 23 | 27 |
| Aspartate aminotransferase (AST, U/L, 29.2–54.8) | 33 | 125 |
| Lactate dehydrogenase (LDH, U/L, 154–353) | 277 | 2072 |
| **Adenovirus load (units)** | | |
| Pharyngeal swab (copies/mL) | $1.36 \times 10^4$ | $7.10 \times 10^8$ |

"/" indicates not done.

Longitudinal profiling of Case 2 revealed a high respiratory HAdV load early in the illness, accompanied by systemic inflammation, coagulation abnormalities, and bacterial co-detection (Figure 4). Although the adenovirus load subsequently increased again during clinical monitoring, repeat type-specific testing was not performed; therefore, whether this increase reflected persistent HAdV-14 detection or another adenovirus type could not be determined. Immune profiling on hospital day 20 showed a reduced natural killer (NK) cell percentage and count and reduced $CD19^+$ cell count, together with increased interferon-alpha (IFN-$\alpha$), interleukin-6 (IL-6), IL-8, and IL-10 levels; immunoglobulin and complement levels were within the corresponding reference ranges (Table 4). During the first 28 days of treatment, respiratory HAdV test results gradually became negative, accompanied by initial clinical stabilization. However, the patient deteriorated again in association

with multi-bacterial infections and pulmonary complications, requiring re-intubation and intensified respiratory support. Brain imaging showed cerebral atrophy and subdural effusion (Figure 3D). Given the preceding cardiopulmonary resuscitation and severe hypoxemia, the neurological injury was clinically attributed to hypoxic–ischemic injury rather than direct central nervous system infection by HAdV-14. Once clinically stable, the patient was transferred from the ICU to the general ward on day 43. Bronchiectasis, neurodevelopmental abnormalities, and global developmental delay (GDD) persisted, and bronchiolitis obliterans (BO) was diagnosed during follow-up.

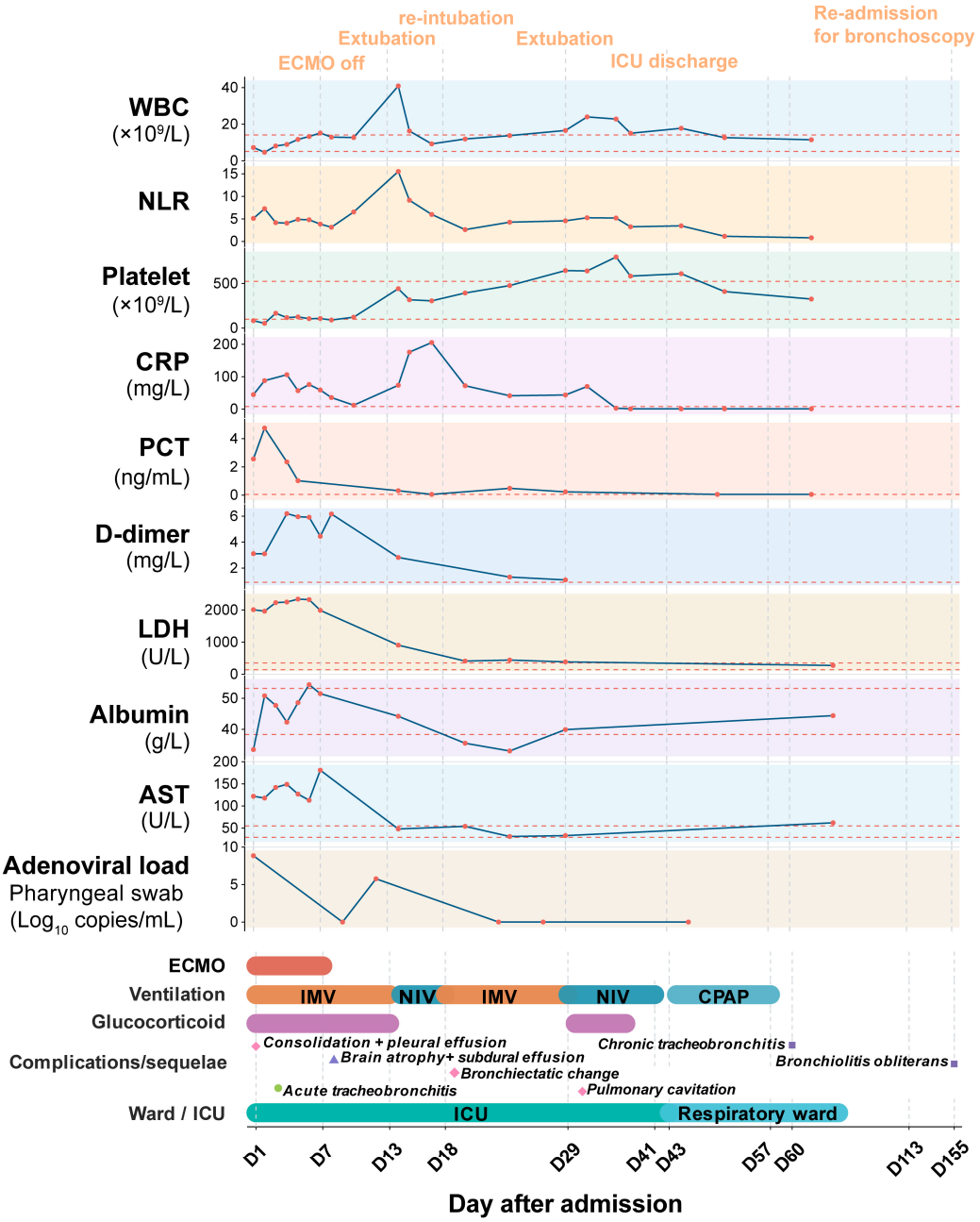



**Figure 4.** Longitudinal clinical course of Case 2. Longitudinal changes in laboratory parameters and respiratory HAdV load are shown together with respiratory support, major therapeutic interventions, complications, sequelae, and ward transfers from admission through follow-up. Red dashed lines indicate the corresponding upper or lower reference limits, where applicable. Abbreviations: AST, aspartate aminotransferase; CPAP, continuous positive airway pressure; CRP, C-reactive protein; ECMO, extracorporeal membrane oxygenation; HAdV, human adenovirus; ICU, intensive care

unit; IMV, invasive mechanical ventilation; LDH, lactate dehydrogenase; NIV, non-invasive ventilation; NLR, neutrophil-to-lymphocyte ratio; PCT, procalcitonin; WBC, white blood cell count.

**Table 4.** Immune profiles of Case 2 on day 20 after admission.

| Items | Indicators | Results | Indicators | Results |
|---|---|---|---|---|
| Lymphocyte subpopulation (unit, reference range) | $CD3^+$ % (%, 39–73) | 79.06 | $CD3^+$ count (cells/μL, 1400–8000) | 2120.21 |
| | $CD3^+$ $CD8^+$ % (%, 11–32) | 27.01 | $CD3^+$ $CD8^+$ count (cells/μL, 400–2300) | 724.30 |
| | $CD3^+$ $CD4^+$ % (%, 25–50) | 50.00 | $CD3^+$ $CD4^+$ count (cells/μL, 900–5500) | 1340.85 |
| | $CD3^+$ $CD4^+$ $CD8^+$ % | 0.13 | $CD3^+$ $CD4^+$ $CD8^+$ count (cells/μL) | 3.59 |
| | $CD3^+$ $CD4^-$ $CD8^-$% | 2.19 | NK count (cells/μL, 100–1400) | 71.83 |
| | NK% (%, 3–16) | 2.68 | $CD19^+$ count (cells/μL, 600–3100) | 486.06 |
| | $CD19^+$ % (%, 7–41) | 18.13 | $CD45^+$ count (cells/μL) | 2681.69 |
| | CD4/CD8 (0.98–1.94) | 1.85 | | |
| Cytokine (units, reference range) | IL-2 (pg/mL, 0–9.80) | 1.50 | IL-10 (pg/mL, 0–4.90) | 143.50 |
| | IL-12p70 (pg/mL, 0–3.40) | 1.93 | TNF-α (pg/mL, 0–5.20) | 3.34 |
| | IFN-α (pg/mL, 0–8.50) | 33.92 | IFN-γ (pg/mL, 0–17.30) | 3.01 |
| | IL-8 (pg/mL, 0–20.60) | 437.91 | IL-17A (pg/mL, 0–14.80) | 3.21 |
| | IL-4 (pg/mL, 0–3.00) | 1.19 | IL-1β (pg/mL, 0–12.4) | 3.08 |
| | IL-5 (pg/mL, 0–3.10) | 0.27 | IL-6 (pg/mL, 0–16.60) | 176.10 |
| Humoral Immunity Indicators (units, reference range) | IgG (g/L, 2.86–16.80) | 14.40 | IgE (IU/mL, 0–165) | 51.00 |
| | IgA (g/L, 0.19–1.75) | 0.48 | Complement C3 (g/L, 0.7–2.06) | 1.13 |
| | IgM (g/L, 0.43–1.63) | 1.39 | Complement C4 (g/L, 0.11–0.61) | 0.28 |

### *3.5. Phylogenetic and Comparative Genomic Analysis of HAdV-14 Isolates with Available Clinical Information*

CQ202503, isolated from the respiratory sample of Case 2, was assembled as a single 34,496-bp coding-complete genome with unresolved termini (Figure S2). Phylogenetic analysis of the whole-genome and the *penton base*, *hexon*, and *fiber* genes consistently assigned CQ202503 to HAdV-14 (P14H14F14), with no evidence of a newly recombinant genotype (Figure S3). In the HAdV-14 whole-genome phylogeny, CQ202503 clustered within the HAdV-14p1-II clade (Figure 5A), consistent with its gene-level phylogenetic placement (Figure S4).

CQ202503 was highly similar to representative HAdV-14p1 strains and showed greater divergence from the historical de Wit prototype (Figure 5B). CQ202503 exhibited high whole-genome nucleotide identity with previously reported Chinese HAdV-14p1 strains, including Beijing 2010 (99.9971%; JN032132.1), GZ01 2010 (99.9855%; JQ824845.1), GZ02 2010 (99.9855%; MF062484.1), and Beijing 2012 (99.9652%; JX892927.2), at comparable alignment sites (Figure 5B; Table S6). Among previously reported Chinese strains, CQ202503 was most similar to Beijing 2010, differing by only one nonsynonymous substitution, and was also closely related to GZ01/GZ02 2010, with four coding-region nucleotide differences involving two amino acid changes. Coding-region differences were distributed across a limited number of genes and did not cluster in the major capsid genes encoding penton base, hexon, or fiber (Figure 5C). Overall, CQ202503 fell within the

known genomic diversity of HAdV-14p1 and showed no marked genomic divergence from previously reported strains, including representative Chinese HAdV-14p1 isolates.

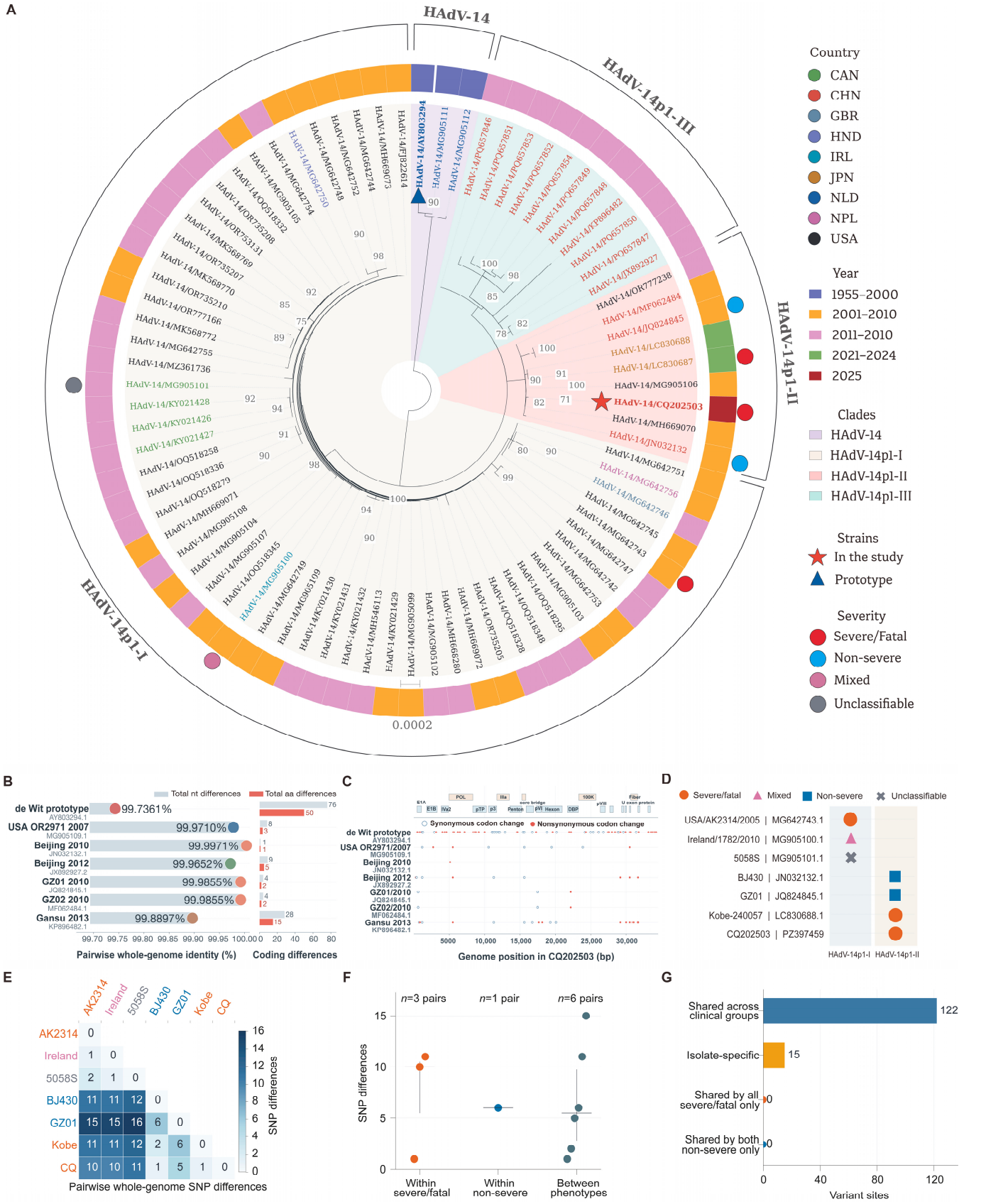



**Figure 5.** Phylogenetic placement and comparative genomic analysis of CQ202503 and HAdV-14 isolates with available pediatric clinical information. (**A**) Maximum-likelihood phylogeny of

CQ202503 and publicly available HAdV-14 whole-genome sequences. The red star indicates CQ202503 and the blue triangle indicates the historical prototype strain de Wit. Tip-label colors indicate countries of origin, the outer ring indicates year groups, and the outermost labels denote the HAdV-14 prototype lineage and HAdV-14p1 clades. Colored circles outside the tree indicate the clinical severity categories of the seven isolates with available pediatric clinical information: severe/fatal, non-severe, mixed, and unclassifiable. (**B**) Pairwise whole-genome identity and coding differences between CQ202503 and selected representative HAdV-14 strains. The left panel shows whole-genome sequence identity, and the right panel summarizes coding differences. (**C**) Genomic distribution of synonymous and nonsynonymous differences between CQ202503 and representative strains across the CQ202503 genome. (**D**) Distribution of the seven clinical-information-linked isolates across the HAdV-14p1-I and HAdV-14p1-II clades. (**E**) Lower-triangular heatmap showing pairwise whole-genome SNP differences among the seven isolates. Numbers in individual cells indicate nucleotide differences between isolate pairs. (**F**) Pairwise SNP differences among the five isolates with unambiguous individual-level binary clinical classifications. Each dot represents one pairwise comparison. Horizontal bars indicate medians, and vertical bars indicate interquartile ranges where applicable. Numbers above each group indicate the numbers of pairwise comparisons. These comparisons are descriptive because pairwise observations are non-independent. (**G**) Distribution of nucleotide variant sites according to their sharing patterns across clinical phenotype categories. Shared across clinical groups indicates variants detected in isolates belonging to more than one clinical category; isolate-specific indicates variants detected in only one isolate. The final two categories indicate variants shared by all three severe/fatal isolates but absent from both non-severe isolates, and variants shared by both non-severe isolates but absent from all severe/fatal isolates, respectively. Abbreviations: CAN, Canada; CHN, China; GBR, Great Britain; HND, Honduras; IRL, Ireland; JPN, Japan; NLD, the Netherlands; NPL, Nepal; USA, United States of America; nt, nucleotide; aa, amino acid.

To further explore the relationship between viral genomic background and clinical phenotype, seven HAdV-14 whole-genome sequence records with available pediatric clinical information were examined. These comprised three severe/fatal isolates (USA/AK2314/2005, Kobe-240057, and CQ202503), two non-severe isolates (BJ430 and GZ01), one mixed-phenotype isolate (Ireland/1782/2010), and one unclassifiable isolate (5058S) (Figure 5A,D). The seven isolates belonged to two established HAdV-14p1 clades. Within HAdV-14p1-I, the isolates included one severe/fatal, one mixed, and one unclassifiable record. Within HAdV-14p1-II, both severe/fatal and non-severe isolates were represented. Thus, severe/fatal isolates occurred in both clades, and no distinct clustering by clinical severity was evident.

Pairwise whole-genome comparisons among the seven isolates showed 1–16 SNP differences across the 21 pairwise comparisons (Figure 5E). The three HAdV-14p1-I isolates differed by 1–2 SNPs, whereas the four HAdV-14p1-II isolates differed by 1–6 SNPs; comparisons between the two clades showed 10–16 SNP differences. In the binary comparison restricted to three severe/fatal and two non-severe isolates, within-group comparisons among severe/fatal isolates showed 1, 10, and 11 SNP differences (n = 3 pairs), whereas the single non-severe pair differed by 6 SNPs (n = 1 pair). Comparisons between severe/fatal and non-severe isolates showed 1–15 SNP differences (n = 6 pairs), with individual values of 1, 2, 5, 6, 11, and 15. The observed distances overlapped across comparison categories, without an apparent separation according to clinical phenotype (Figure 5F).

Prototype-relative variant analysis identified 137 distinct nucleotide variant records across the seven isolates. Of these, 122 were shared across more than one clinical phenotype category, whereas 15 were isolate-specific. No variant was shared by all three severe/fatal isolates while being absent from both non-severe isolates. Similarly, no variant

was shared by both non-severe isolates while being absent from all severe/fatal isolates (Figure 5G). Overall, the available genomic data did not reveal an obvious phenotype-specific consensus variant pattern distinguishing severe/fatal from non-severe pediatric HAdV-14 infections.

## 4. Discussion

The principal finding of this study was that reported pediatric HAdV-14 infections were not uniformly severe across different case-ascertainment settings. Across the available pediatric reports, the clinical spectrum extended from uncomplicated febrile respiratory illness and upper respiratory tract infection to severe pneumonia, ARDS, ECMO-supported critical illness, death, and long-term pulmonary or neurological sequelae. Notably, cases identified through outbreak investigations were predominantly non-severe: affected children most commonly presented with fever and upper respiratory tract symptoms, and none of the 111 outbreak-associated cases included in our analysis were classified as severe or fatal. Similar heterogeneity has been observed in adults. During a large US military outbreak, HAdV-14 infection was common, but most infections identified through cohort surveillance were mild and no infected cohort member required hospitalization [27]; among military trainees with pneumonia, HAdV-14 positivity was not associated with greater pneumonia severity, ICU admission, longer hospitalization, or 30-day mortality [5]. Taken together, these observations indicate that HAdV-14 has the potential to cause life-threatening disease, but the severe clinical presentations emphasized in sporadic reports do not appear to characterize all settings in which pediatric HAdV-14 infection has been systematically identified.

The marked difference in reported severity across case-ascertainment settings raises the possibility that the historical perception of HAdV-14 as a cause of severe disease has been influenced, at least in part, by case detection and reporting practices. Outbreak investigations systematically screen a defined population and therefore capture the broader spectrum of infection, including uncomplicated febrile illnesses. Sporadic reports, in contrast, may preferentially arise from unusual, complicated, or life-threatening cases that prompt type-specific testing and publication. This potential bias has previously been recognized in HAdV-14 surveillance: the first reported Irish HAdV-14p1 series included three deaths among nine cases, yet the investigators explicitly noted that hospital-diagnosed cases might represent the severe end of the disease spectrum [17]. More generally, adenovirus typing is not routinely performed in clinical practice and is disproportionately undertaken during severe infections and outbreak investigations [8]. Our pediatric findings are consistent with the potential influence of this ascertainment structure: severe and fatal cases were concentrated in sporadic reports, whereas outbreak-associated cases were predominantly non-severe. Accordingly, the high proportion of severe cases in the published sporadic literature should not be interpreted as an estimate of the population-level probability of severe HAdV-14 infection. Importantly, however, outbreak-associated and sporadic cases also differed in age, host characteristics, and epidemiological context; therefore, the observed difference in severity cannot be attributed to ascertainment or reporting alone. Rather, selective case capture may be one contributor to the severity profile of published pediatric cases. Furthermore, most published cases originated from China, which may limit the generalizability of the observed clinical spectrum to other populations.

Among the clinical variables available for sporadic cases, adverse premorbid conditions were reported more frequently among severe/fatal cases than among non-severe cases. They were present in more than half of the severe/fatal cases but in only a small minority of non-severe cases, whereas age, sex, season and co-detection were not significantly associated with disease severity in the available data. The reported premorbid

conditions were heterogeneous—including prematurity, recurrent lower respiratory tract disease, developmental abnormalities, immune deficiency, and suspected pathogenic chromosomal abnormalities—suggesting that broader premorbid vulnerability, rather than any single specific comorbidity, may be clinically relevant. Studies of previous adult HAdV-14 outbreaks have also reported associations between underlying health conditions and severe outcomes [4,28], and broader pediatric adenovirus studies have highlighted the potential contribution of pre-existing conditions to disease severity [29]. Although these limited case-level data do not permit assessment of independent risk factors, the observed difference suggests a possible association between premorbid host vulnerability and severe/fatal outcomes. This finding should be considered as hypothesis-generating and requires validation in larger studies with standardized documentation of underlying health conditions.

Premorbid vulnerability, however, does not explain all severe disease. The critically ill child described in detail in this study had no established immunodeficiency, although a suspected pathogenic copy-number variation (CNV) was identified. His clinical course illustrates the complex and evolving factors that may accompany critical HAdV-14 infection. During the acute phase, a high respiratory HAdV load coincided with marked inflammatory and coagulation abnormalities and altered lymphocyte subsets and cytokine responses; multiple bacterial infections and major pulmonary and neurological complications developed later. Importantly, clinical deterioration and long-term pulmonary and neurological sequelae persisted after respiratory HAdV became undetectable, with hypoxic–ischemic injury and subsequent multidrug-resistant bacterial infection considered important contributors to the later clinical course. These findings are consistent with severe adenovirus disease as a dynamic process rather than a consequence of viral type alone. High viral burden and dysregulated inflammatory responses may contribute to tissue injury, whereas secondary bacterial infection and prolonged critical illness may further amplify pulmonary damage and long-term sequelae. HAdV-14 detection alone should not be regarded as a marker of poor prognosis. Together, these observations suggest a clinically relevant distinction between pre-existing host vulnerability, which may predispose to severe disease, and acute disease-amplifying factors, including high viral burden, inflammatory dysregulation, secondary infection, and critical-care complications.

The genomic findings further suggest that viral lineage alone is unlikely to account for the marked variability in clinical severity. The severe-case isolate CQ202503 belonged to the established HAdV-14p1-II lineage and was highly similar to previously characterized HAdV-14p1 strains. The exploratory genomic comparisons did not reveal a clear clade-level or consensus-variant pattern corresponding to clinical severity among the pediatric HAdV-14 isolates examined. This observation is consistent with the study by Houng et al., in which HAdV-14 isolates obtained from mild respiratory infections and fatal pneumonia showed very high genomic similarity and no clear genomic determinant explaining the marked difference in clinical outcome [6]. Collectively, these observations do not identify a clear consensus-level genomic signature that distinguishes severe/fatal from non-severe pediatric HAdV-14 infections. Nevertheless, the small number of isolates with unambiguous individual-level clinical classifications limits the strength of this inference, and a contribution of viral genetic variation to disease severity cannot be excluded. HAdV-14p1 is genetically distinct from the historical prototype and may have arisen through recombination, while ongoing microevolution within the HAdV-14p1 lineage has also been documented [2,26]. However, available genomic comparisons have not identified viral variants that clearly account for differences in clinical severity [6], and the limited number of phenotype-linked genomes means that more subtle genetic determinants cannot yet be excluded.

These findings have direct implications for the clinical interpretation and surveillance of HAdV-14. First, identification of HAdV-14 should alert clinicians to the potential for critical respiratory disease but should not imply that severe progression is inevitable. In children with HAdV-14 infection, premorbid host vulnerability and the evolving clinical course should be considered alongside viral type when assessing disease severity. Second, type-specific HAdV-14 surveillance should extend beyond critically ill patients. Non-severe outbreak-associated infections constitute an important part of the reported clinical spectrum, and their inclusion is necessary to better characterize transmission and the population-level distribution of disease severity. Prospective multicenter surveillance with systematic adenovirus typing would therefore provide a more representative assessment than surveillance based predominantly on selectively typed severe cases. Finally, severe or fatal pediatric HAdV-14 infections remain particularly valuable for combined clinical and genomic investigation because linking host characteristics with viral burden, immune responses, and viral genomes may help clarify why some children develop life-threatening disease.

Several limitations should be considered. First, the available pediatric HAdV-14 literature remains limited and heterogeneous, with incomplete reporting of clinical variables and potential publication and ascertainment biases. Because standardized physiological parameters were unavailable for most literature-derived cases, we used an operational severity classification based on extractable clinical diagnoses, treatment interventions, and outcomes rather than applying a standardized CAP severity scoring system uniformly. This approach may have introduced some degree of misclassification. Second, outbreak-associated and sporadic cases originated from different populations, age groups, study periods, and epidemiological settings; therefore, the observed differences in disease severity cannot be attributed to case ascertainment alone, nor can these data be used to estimate the population-level risk of severe HAdV-14 infection. Third, co-infection testing and reporting were incomplete, and the pathogen panels varied across studies. Consequently, the potential contributions of viral co-detection and secondary bacterial infections could not be adequately assessed, and severe outcomes cannot be confidently attributed to HAdV-14 alone. Fourth, the exploratory association between adverse premorbid conditions and severe/fatal disease was based on a limited number of cases with variable data completeness, and no multivariable risk modelling was feasible. Fifth, only two local cases were available, and genomic characterization was performed for a single severe-case isolate. Although the expanded genomic analysis included seven HAdV-14 whole-genome sequence records with available pediatric clinical information, only five had unambiguous individual-level binary severity classifications. These limited and heterogeneous clinical-genomic data limit the strength of genotype–phenotype inference. Finally, the retrospective, literature-based nature of the case-level synthesis cannot substitute for prospective cohorts with systematic adenovirus typing. Prospective multicenter studies are needed to validate the observed severity patterns and host-related associations.

## 5. Conclusions

In the available pediatric evidence, the reported severity of HAdV-14 infection differed substantially across case-ascertainment settings, with outbreak-associated cases showing a less severe clinical profile than sporadically reported cases. Adverse premorbid conditions were reported more frequently among severe/fatal sporadic cases, suggesting that host vulnerability may contribute to the marked heterogeneity in clinical outcomes. Comparative analysis of seven HAdV-14 genomes with pediatric clinical information did not reveal an obvious clade-level or consensus-variant pattern corresponding to clinical severity; however, the limited number of isolates with unambiguous individual-level

phenotypes precludes definitive conclusions about genotype–phenotype relationships. Together, these findings suggest that the severe phenotype historically associated with pediatric HAdV-14 should be interpreted in the context of both case ascertainment and host characteristics, and warrant validation through prospective studies with systematic adenovirus typing.

**Supplementary Materials:** The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/v18121343/s1: Supplementary Materials and Methods; Figure S1: PRISMA-like flow diagram of the literature search and selection process; Figure S2: Genome structure, coding-region integrity, and sequencing depth of CQ202503; Figure S3: Broad adenovirus typing of CQ202503 based on whole-genome, *penton base*, *hexon*, and *fiber* phylogenies; Figure S4: Phylogenetic placement of CQ202503 based on HAdV-14 *penton base*, *hexon*, and *fiber* genes; Table S1: Risk-of-bias assessment of included case control studies; Table S2: Risk-of-bias assessment of included case reports; Table S3: Risk-of-bias assessment of included cross-sectional studies; Table S4: Risk-of-bias assessment of included case series; Table S5: Risk-of-bias assessment of included prevalence studies; Table S6: HAdV-14 reference sequences used for genomic and phylogenetic analyses; Table S7: Reported outbreak/cluster-associated pediatric HAdV-14 cases; Table S8: Reported sporadic pediatric HAdV-14 cases. Reference [30] is cited in the supplementary materials.

**Author Contributions:** Conceptualization, N.Z., Y.F. and E.L.; methodology, J.C.; formal analysis, J.C.; investigation, J.C., Y.L. and Z.Y.; resources, F.Y., Y.Y., H.Q., N.Z. and Y.F.; data curation, J.C., F.Y., Y.Y., H.Q., N.Z. and Y.F.; writing—original draft preparation, J.C.; writing—review and editing, E.L., Y.W., N.Z. and Y.F.; visualization, J.C.; supervision, N.Z., Y.F. and E.L. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research was funded by Major Project of Guangzhou National Laboratory grant number GZNL2023A01004.

**Institutional Review Board Statement:** The study was conducted in accordance with the Declaration of Helsinki, and approved by the Ethics Committee of Children's Hospital of Chongqing Medical University (reference number: 2024 Ethics Review [Research] No. 217-1).

**Informed Consent Statement:** Written informed consent was obtained from the parents or legal guardians of the two local pediatric cases for the use and publication of clinical data and residual clinical specimens.

**Data Availability Statement:** The raw sequencing data generated in this study are publicly available in the NCBI Sequence Read Archive (SRA) under BioProject accession number PRJNA1463702 and SRA accession number SRR38451428. The HAdV-14 genome sequence generated in this study has been deposited in GenBank under accession number PZ397459. Publicly available viral sequences used for comparative genomic and phylogenetic analyses were obtained from GenBank, with accession numbers provided in Table S6. Individual-level clinical data from the local pediatric cases are not publicly available because of patient privacy and ethical restrictions but may be available from the corresponding authors upon reasonable request, subject to applicable ethical and institutional requirements.

**Acknowledgments:** We sincerely thank the patients and their caregivers for their participation. We also thank the staff of the Department of Respiratory Medicine and the Intensive Care Unit of Children's Hospital of Chongqing Medical University for their assistance with nasopharyngeal aspirate collection and clinical care.

**Conflicts of Interest:** The authors declare no conflicts of interest.

## Abbreviations

The following abbreviations are used in this manuscript:

| | |
|---|---|
| ALT | Alanine aminotransferase |
| ARD | Acute respiratory disease |
| ARDS | Acute respiratory distress syndrome |
| AST | Aspartate aminotransferase |
| BAM | Binary alignment/map |
| BGI | Beijing Genomics Institute |
| BLAST | Basic Local Alignment Search Tool |
| BO | Bronchiolitis obliterans |
| BWA-MEM | Burrows–Wheeler Aligner maximal exact matches |
| CAN | Canada |
| CDC | Centers for Disease Control and Prevention |
| CDS | Coding sequence |
| CHN | China |
| CI | Confidence interval |
| CNV/CNVs | Copy number variation/copy number variations |
| CPAP | Continuous positive airway pressure |
| CRP | C-reactive protein |
| CT | Computed tomography |
| DNBSEQ | DNA nanoball sequencing |
| ECMO | Extracorporeal membrane oxygenation |
| EPS | Encapsulated PostScript |
| FASTA | FASTA sequence format |
| FRI | Febrile respiratory illness |
| GBR | Great Britain |
| GDD | Global developmental delay |
| GTR + G | General time-reversible model with gamma-distributed rate heterogeneity |
| HAdV | Human adenovirus |
| HAdV-14 | Human adenovirus type 14 |
| HAdV-14p | Prototype human adenovirus type 14 |
| HAdV-14p1 | Human adenovirus type 14p1 variant |
| HIBD | Hypoxic–ischemic brain damage |
| HND | Honduras |
| HRV | Human rhinovirus |
| ICU | Intensive care unit |
| IFN | Interferon |
| IFN-$\alpha$ | Interferon-alpha |
| IFN-$\gamma$ | Interferon-gamma |
| IgA | Immunoglobulin A |
| IgE | Immunoglobulin E |
| IgG | Immunoglobulin G |
| IgM | Immunoglobulin M |
| IL | Interleukin |
| IMV | Invasive mechanical ventilation |
| IQ-TREE | Intelligent tree; phylogenetic software for maximum-likelihood tree inference |
| IRL | Ireland |
| IVIG | Intravenous immunoglobulin |
| JBI | Joanna Briggs Institute |
| JPN | Japan |

| | |
|---|---|
| LDH | Lactate dehydrogenase |
| MAFFT | Multiple alignment using fast Fourier transform |
| mNGS | Metagenomic next-generation sequencing |
| NCBI | National Center for Biotechnology Information |
| NIV | Non-invasive ventilation |
| NK cell | Natural killer cell |
| NLD | The Netherlands |
| NLR | Neutrophil-to-lymphocyte ratio |
| NPL | Nepal |
| OR | Odds ratio |
| ORF | Open reading frame |
| PCR | Polymerase chain reaction |
| PCT | Procalcitonin |
| PICU | Pediatric intensive care unit |
| Q1 | First quartile |
| Q3 | Third quartile |
| SAMtools | Sequence Alignment/Map tools |
| SNP | Single-nucleotide polymorphism |
| TNF-$\alpha$ | Tumor necrosis factor-alpha |

## References


1. Stopczynski, T.; Probst, V.; Gailani, A.; Amarin, J.Z.; Hamdan, O.; Hayek, H.; Stewart, L.S.; Rahman, H.K.; Selvarangan, R.; Schuster, J.E.; et al. Adenovirus Types in US Children Hospitalized or Seen in the Emergency Department With Acute Respiratory Illness, 2016–2019. *Open Forum Infect. Dis.* **2025**, *12*, ofaf692. https://doi.org/10.1093/ofid/ofaf692.
2. Kajon, A.E.; Lu, X.; Erdman, D.D.; Louie, J.; Schnurr, D.; George, K.S.; Koopmans, M.P.; Allibhai, T.; Metzgar, D. Molecular Epidemiology and Brief History of Emerging Adenovirus 14-Associated Respiratory Disease in the United States. *J. Infect. Dis.* **2010**, *202*, 93–103. https://doi.org/10.1086/653083.
3. Centers for Disease Control and Prevention (CDC) Acute Respiratory Disease Associated with Adenovirus Serotype 14--Four States, 2006–2007. *MMWR Morb. Mortal. Wkly. Rep.* **2007**, *56*, 1181–1184.
4. Lewis, P.F.; Schmidt, M.A.; Lu, X.; Erdman, D.D.; Campbell, M.; Thomas, A.; Cieslak, P.R.; Grenz, L.D.; Tsaknardis, L.; Gleaves, C.; et al. A Community-Based Outbreak of Severe Respiratory Illness Caused by Human Adenovirus Serotype 14. *J. Infect. Dis.* **2009**, *199*, 1427–1434. https://doi.org/10.1086/598521.
5. Vento, T.J.; Prakash, V.; Murray, C.K.; Brosch, L.C.; Tchandja, J.B.; Cogburn, C.; Yun, H.C. Pneumonia in Military Trainees: A Comparison Study Based on Adenovirus Serotype 14 Infection. *J. Infect. Dis.* **2011**, *203*, 1388–1395. https://doi.org/10.1093/infdis/jir040.
6. Houng, H.-S.H.; Gong, H.; Kajon, A.E.; Jones, M.S.; Kuschner, R.A.; Lyons, A.; Lott, L.; Lin, K.-H.; Metzgar, D. Genome Sequences of Human Adenovirus 14 Isolates from Mild Respiratory Cases and a Fatal Pneumonia, Isolated during 2006-2007 Epidemics in North America. *Respir. Res.* **2010**, *11*, 116. https://doi.org/10.1186/1465-9921-11-116.
7. Laham, F.R.; Jewell, A.M.; Schoonover, S.L.; Demmler, G.J.; Piedra, P.A. The Search for Adenovirus 14 in Children in Houston, Texas. *Pediatr. Infect. Dis. J.* **2008**, *27*, 653–654.
8. Abdirizak, F.; Winn, A.K.; Parikh, R.; Scobie, H.M.; Lu, X.; Vega, E.; Almendares, O.; Kirking, H.L.; Rose, E.B.; Silk, B.J. Surveillance of Human Adenovirus Types and the Impact of the COVID-19 Pandemic on Reporting—United States, 2017–2023. *Morb. Mortal. Wkly. Rep.* **2024**, *73*, 1136–1141. https://doi.org/10.15585/mmwr.mm7350a1.
9. Expert Writing Group of Technical Guidelines for Prevention and Control of Human Adenovirus Respiratory Infection. Technical guidelines for prevention and control of human adenovirus respiratory infection (2019 edition). *Chin. J. Prev. Med.* **2019**, *53*, 1088–1093. https://doi.org/10.3760/cma.j.issn.0253-9624.2019.11.003.
10. Subspecialty Group of Respiratory, the Society of Pediatrics, Chinese Medical Association; Editorial Board, Chinese Journal of Pediatrics; China Medicine Education Association Committee on Pediatrics. Guidelines for the Management of Community-Acquired Pneumonia in Children (2024 Revision). *Chin. J. Pediatr.* **2024**, *62*, 920–930. https://doi.org/10.3760/cma.j.cn112140-20240728-00523. (In Chinese).

11. Huang, G.; Yu, D.; Zhu, Z.; Zhao, H.; Wang, P.; Gray, G.C.; Meng, L.; Xu, W. Outbreak of Febrile Respiratory Illness Associated with Human Adenovirus Type 14p1 in Gansu Province, China. *Influenza Other Respir. Viruses* **2013**, *7*, 1048–1054. https://doi.org/10.1111/irv.12118.
12. Ying, L.; Qing-Feng, S.; Ye, C.; Ying, Q. Investigation on an Outbreak of Acute Respiratory Infection in Shenyang. *Mod. Prev. Med.* **2014**, *41*, 3600–3602. (In Chinese)
13. Wenwen, Z.; Weiwei, W.; Chuanwu, S.; Jun, B.; Yangguang, D.; Jing, T. Analysis of Two Outbreaks of Respiratory Infection Caused by Adenovirus. *Chin. J. Exp. Clin. Virol.* **2018**, *32*, 289–291. https://doi.org/10.3760/cma.j.issn.1003-9279.2018.03.016. (In Chinese)
14. Girouard, G.; Garceau, R.; Thibault, L.; Oussedik, Y.; Bastien, N.; Li, Y. Adenovirus Serotype 14 Infection, New Brunswick, Canada, 2011. *Emerg. Infect. Dis.* **2013**, *19*, 119–122. https://doi.org/10.3201/eid1901.120423.
15. Chen, H.-L.; Chiou, S.-S.; Hsiao, H.-P.; Lin, Y.-C.; Lin, K.-H.; Jong, Y.-J. Respiratory Adenoviral Infections in Children: A Study of Hospitalized Cases in Southern Taiwan in 2001–2002. *J. Trop. Pediatr.* **2004**, *50*, 279–284. https://doi.org/10.1093/tropej/50.5.279.
16. Louie, J.K.; Kajon, A.E.; Holodniy, M.; Guardia-LaBar, L.; Lee, B.; Petru, A.M.; Hacker, J.K.; Schnurr, D.P. Severe Pneumonia Due to Adenovirus Serotype 14: A New Respiratory Threat? *Clin. Infect. Dis. Off. Publ. Infect. Dis. Soc. Am.* **2008**, *46*, 421–425. https://doi.org/10.1086/525261.
17. O'Flanagan, D.; O'Donnell, J.; Domegan, L.; Fitzpatrick, F.; Connell, J.; Coughlan, S.; De Gascun, C.; Carr, M.J. First Reported Cases of Human Adenovirus Serotype 14p1 Infection, Ireland, October 2009 to July 2010. *Euro Surveill. Bull. Eur. Sur Mal. Transm. Eur. Commun. Dis. Bull.* **2011**, *16*, 19801.
18. Carr, M.J.; Kajon, A.E.; Lu, X.; Dunford, L.; O'Reilly, P.; Holder, P.; De Gascun, C.F.; Coughlan, S.; Connell, J.; Erdman, D.D.; et al. Deaths Associated with Human Adenovirus-14p1 Infections, Europe, 2009–2010. *Emerg. Infect. Dis.* **2011**, *17*, 1402–1408. https://doi.org/10.3201/eid1708.101760.
19. Tang, L.; An, J.; Xie, Z.; Dehghan, S.; Seto, D.; Xu, W.; Ji, Y. Genome and Bioinformatic Analysis of a HAdV-B14p1 Virus Isolated from a Baby with Pneumonia in Beijing, China. *PLoS ONE* **2013**, *8*, e60345. https://doi.org/10.1371/journal.pone.0060345.
20. Mizuno, S.; Tanimoto, Y.; Mori, A.; Fuseya, T.; Ishida, Y.; Nishiyama, M.; Maruyama, A.; Kasai, M. Acute Encephalopathy Associated with Human Adenovirus Type 14 Infection in 7-Year-Old Girl, Japan. *Emerg. Infect. Dis.* **2025**, *31*, 377–379. https://doi.org/10.3201/eid3102.241168.
21. Jirong, L.; Wenyong, F. Three Cases of Upper and Lower Respiratory Tract Infections Caused by Adenovirus Type 14. *Chin. J. Pediatr.* **1990**, *28*, 372. https://doi.org/10.3760/cma.j.issn.0578-1310.1990.06.142. (In Chinese)
22. Zhang, Q.; Seto, D.; Zhao, S.; Zhu, L.; Zhao, W.; Wan, C. Genome Sequence of the First Human Adenovirus Type 14 Isolated in China. *J. Virol.* **2012**, *86*, 7019–7020. https://doi.org/10.1128/JVI.00814-12.
23. Xiaofang, L.; Bing, Z.; Zhaojun, D.; Qin, L.; Zhiping, X. Molecular Epidemiological Investigation of Adenovirus in Hospitalized Children Associated with Acute Lower Respiratory Infection in Changsha from 2013-2014. *Chin. J. Exp. Clin. Virol.* **2015**, *29*, 505–509. https://doi.org/10.3760/cma.j.issn.1003-9279.2015.06.010. (In Chinese)
24. Mengchan, C.; Jun, H.; Xianxiang, L.; Siqing, X. Identification of Adenovirus Epidemic Strains and Analysis of Hexon Gene Characteristics in Anqing Area from 2013 to 2015. *Chin. J. Microbiol. Immunol.* **2018**, *38*, 855–861. https://doi.org/10.3760/cma.j.issn.0254-5101.2018.11.009. (In Chinese)
25. Wenwen, Z.; Weiwei, W.; Yanping, X.; Yangguang, D.; Liuying, T.; Jing, T. Epidemiological Characteristics and Genotypes of Human Adenovirus among Children in Xuzhou from 2013 to 2017. *Int. J. Virol.* **2018**, *25*, 315–318. https://doi.org/10.3760/cma.j.issn.1673-4092.2018.05.008. (In Chinese)
26. Jin, Y.; Wang, J.; Mao, N.; Zhu, C.; Zhou, H.; He, W.; Ding, H.; Xia, B.; Cui, A.; Zhang, Y.; et al. Genomic Insights into the 2024 HAdV-14p1 Strain in China: Traced to Early Prevalent Strains. *China CDC Wkly.* **2026**, *8*, 195–200. https://doi.org/10.46234/ccdcw2026.033.
27. Tate, J.E.; Bunning, M.L.; Lott, L.; Lu, X.; Su, J.; Metzgar, D.; Brosch, L.; Panozzo, C.A.; Marconi, V.C.; Faix, D.J.; et al. Outbreak of Severe Respiratory Disease Associated with Emergent Human Adenovirus Serotype 14 at a US Air Force Training Facility in 2007. *J. Infect. Dis.* **2009**, *199*, 1419–1426. https://doi.org/10.1086/598520.
28. Centers for Disease Control and Prevention (CDC). Outbreak of Adenovirus 14 Respiratory Illness—Prince of Wales Island, Alaska, 2008. *MMWR Morb. Mortal. Wkly. Rep.* **2010**, *59*, 6–10.

29. Aykac, K.; Kuruc, A.I.; Demir, O.O.; Karatas, E.D.; Ugur, F.; Tuychiboeva, G.; Cengiz, A.B.; Aydin, M.; Ozsurekci, Y. Pediatric Adenovirus Infections: 10-Year Clinical Spectrum and Predictors of Severe Disease with Emphasis on Comorbidities and Coinfections. *Eur. J. Pediatr.* **2026**, *185*, 296. https://doi.org/10.1007/s00431-026-06952-0.
30. Xu, W.; McDonough, M.C.; Erdman, D.D. Species-Specific Identification of Human Adenoviruses by a Multiplex PCR Assay. *J Clin. Microbiol* **2000**, *38*, 4114–4120. https://doi.org/10.1128/JCM.38.11.4114-4120.2000.

**Disclaimer/Publisher's Note:** The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.