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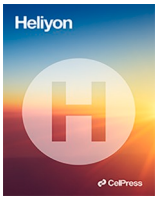

Case report

# Coinfection of *Cytomegalovirus*, *Pneumocystis jirovecii pneumonia*, COVID-19, and *Mycobacterium colombiense* in an AIDS patient: A case report

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ABSTRACT

We present an AIDS patient coinfected with *Cytomegalovirus, Pneumocystis jirovecii pneumonia,* nontuberculous mycobacteria, and COVID-19, who finally recovered from the coinfection. The 36-year-old man had two hospitalizations. In the first hospitalization, the patient was diagnosed with *Cytomegalovirus, Pneumocystis jirovecii pneumonia*, HIV, and COVID-19 quickly and accurately, and the corresponding treatment worked well. The second hospitalization can be divided into four stages: (1) Persistent fever period; (2) Persistent fever and Pulmonary Progression; (3) ICU period; and (4) Pneumothorax period. During the second hospitalization, the diagnosis of Mycobacterium colombiense was hard because the NGS, acid-fast bacilli, and culture of vomit, sputum, and bronchoalveolar lavage fluid were all negative. Still, we detected acid-fast bacilli in the blood mycobacterium culture. In conclusion, we report a severe pneumonia AIDS patient coinfected with *Cytomegalovirus, Pneumocystis jirovecii pneumonia*, COVID-19, and Mycobacterium colombiense who finally recovered from the disease. Nontuberculous mycobacteria infection is common in HIV patients, but bronchoalveolar lavage fluid NGS cannot identify nontuberculous mycobacteria in our report. Traditional blood culture was useful in detecting acid-fast bacilli in our study and then detecting the pathogens with NGS. Combining traditional microbial culture and emerging rapid NGS methods is more conducive to clinical diagnosis and treatment.

## 1. Introduction

Human immunodeficiency virus-1 (HIV-1) has infected approximately 84 million people worldwide and caused nearly 40 million deaths [1] over the past 40 years. HIV attacks the human immune system, destroying CD4 cells and weakening a person's immunity to opportunistic infections, such as tuberculosis (TB), *Cytomegalovirus* (CMV), *Pneumocystis jirovecii pneumonia* (PJP), nontuberculous mycobacteria (NTB), and some cancers [2].

After the first reported coronavirus disease 2019 (COVID-19) in December 2019 [3], COVID-19 swept the world [4–7]. Although 760 million cases and 6.9 million deaths have been reported worldwide, COVID-19 is now an established and ongoing health issue that no longer constitutes a public health emergency of international concern [8]. China has taken a relaxed policy to prevent COVID-19, and COVID-19 is easy for ordinary people to access. Research has found that people living with HIV are not protected from COVID-19,

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and HIV-related immunosuppression may increase the risk of severe COVID-19 [9]. People living with HIV also have a high risk of contracting COVID-19.

*Mycobacterium colombiense (M. Colombiense)* is a new member of the *Mycobacterium avium complex (MAC)*, which was first isolated in 2006 from four Colombian HIV patients [10]. *M. Colombiense* is a rare disease for health populations, but immunocompromised people such as people living with HIV [11] and organ transplant recipients [12] are at greater risk for this pathogen. *M. Colombiense* infection has been reported worldwide, including Canada [13], Brazil [14], Portugal [11], China [15,16], and Japan [17]. In our study, we present an HIV patient coinfected with CMV, PJP, COVID-19, and *M. Colombiense* who finally recovered from the coinfection. To our knowledge, this is the first reported HIV case coinfected with CMV, PJP, COVID-19, and *M. Colombiense* in the world.

## 2. Case presentation

A 36-year-old Han-Chinese man was admitted due to a fever with abdominal pain for two months. The patient developed a fever two months ago, with a maximum temperature of 39 °C accompanied by abdominal pain and anorexia. The patient had severe abdominal pain accompanied by chest pain and fever a month ago and then went to the local hospital for a chest CT scan, which showed scattered glass-like high-density shadows in both lungs. The gastroscopy pathology from the biology was considered a

**Table 1**
$CD4^+$, CD4+/CD8+ ratio, HIV viral load, CMV viral load, and BMI results for the patient at different times.

| Variable | Results |
|---|---|
| $CD4^+$ T cell (/ul); CD4+/CD8+ ratio | |
| 20230530 | 95; 0.11 |
| 20230531 | 96; 0.09 |
| 20230712 | 20; 0.37 |
| 20230722 | 57; 0.79 |
| 20230913 | 160; 0.49 |
| 20231017 (First-month follow-up) | 195; 0.51 |
| 20231114 (Second-month follow-up) | 199; 0.45 |
| 20231214 (Third-month follow-up) | 124; 0.41 |
| 20240117 (Fourth-month follow-up) | 129; 0.33 |
| 20240220 (Fifth-month follow-up) | 140; 0.42 |
| HIV viral load (copies/ml) | |
| 20230530 | 1.44*10^5 |
| 20230713 | 2.77*10^2 |
| 20231017 (First-month follow-up) | <60 |
| 20231114 (Second-month follow-up) | 1.56*10^2 |
| 20231214 (Third-month follow-up) | <60 |
| 20240118 (Fourth-month follow-up) | 1.24*10^2 |
| 20240220 (Fifth-month follow-up) | <60 |
| CMV viral load (copies/ml) | |
| 20230530 (Serum) | 3.16*10^3 |
| 20230601 (Urine) | 9.31*10^5 |
| 20230608 (BALF) | 6.20*10^5 |
| 20230706 (Serum) | <500.00 |
| 20230706 (Urine) | 9.89*10^3 |
| 20230711 (Serum) | <500 |
| 20230713 (Serum) | <500 |
| 20230713 (Urine) | 2.12*10^3 |
| 20230914 (Serum) | <500 |
| 20230914 (Urine) | <500 |
| 20231017 (First-month follow-up, Serum) | <500 |
| 20231019 (First-month follow-up, Urine) | 1.33*10^4 |
| 20231114 (Second-month follow-up, Serum) | <500 |
| 20231116 (Second-month follow-up, Urine) | 5.59*10^3 |
| 20231214 (Third-month follow-up, Serum) | <500 |
| 20231219 (Third-month follow-up, Urine) | <500 |
| 20240118 (Fourth-month follow-up, Serum) | <500 |
| 20240118 (Fourth-month follow-up, Urine) | <500 |
| 20240220 (Fifth-month follow-up, Serum) | <500 |
| 20240220 (Fifth-month follow-up, Urine) | <500 |
| BMI ($kg/m^2$) | |
| 20230530 (First hospitalization admission) | 16.85 (47/1.67 ^ 2) |
| 20230917 (Second hospitalization discharge) | 12.55 (35/1.67^2) |
| 20231018(First-month follow-up) | 14.34 (40/1.67^2) |
| 20231114 (Second-month follow-up) | 17.93 (50/1.67^2) |
| 20231214 (Third-month follow-up) | 18.29 (51/1.67^2) |
| 20240117 (Fourth-month follow-up) | 18.65 (52/1.67^2) |
| 20240219 (Fifth-month follow-up) | 19.00 (53/1.67^2) |

cytomegalovirus infection at the local hospital. Then the patient went to the Second Affiliated Hospital of Zhejiang University for a follow-up gastroscopy, and the results showed multiple ulcers in the stomach and colon. The patient has been transferred to Hangzhou Xixi Hospital due to a positive initial screening for HIV antibodies.

The first hospitalization of the patient was on May 29. The patient was infected with HIV through homosexual behavior. After examination, the patient had AIDS retinopathy in both eyes, with a BMI of 16.85 kg/m$^2$ (47/1.67 ^ 2); NRS2002 score [18] for nutritional risk: 5 points (severe malnutrition); blood gas analysis indicated that the partial arterial oxygen partial pressure (PaO2) was 71.2 mmHg [reference range: 88–108 mmHg], and the patient was positive for SARS-CoV-2 (Table 1). The patient's lung CT showed diffuse infection in both lungs (Fig. 3A). CD4 + T cell count (absolute value) of 95 cells/μL [reference range: 410–1590 cells/μL], CD4/CD8 ratio was 0.11 (Table 1); Metagenomic next generation sequencing (mNGS) using the MGISEQ-200 gene sequencer for bronchoalveolar lavage fluid (BALF) showed that the sequence reads of Pneumocystis jirovecii were 207910, and the sequence reads of CMV were 2399. The patient had no HBV or HCV infection. The treatment plan was methylprednisolone for anti-inflammatory purposes, ganciclovir against CMV, sulfamethoxazole (SMZ) against PJP, and Paxlovid or Molnupiravir against COVID-19 (Fig. 1). The patient started antiviral treatment against HIV with Tenofovir (TDF) + Lamivudine (3TC) + Dolutegravir (DTG) on June 9, and the treatment plan has been ongoing. After a period of treatment, the patient's condition improved, and he was discharged on June 15th (Fig. 1). The detailed dosage and usage for the drugs can be seen in Supplementary Table 1.

The patient was secondly admitted to Hangzhou Xixi Hospital on 26 June for a fever of two days, and the patient was diagnosed as immune reconstitution inflammatory syndrome (IRIS). The second hospitalization can be divided into four stages: (1) Persistent fever period (20230626–20230702): The patient's lung CT showed a better improvement compared to the previous CT result in both lungs (Fig. 3B) at the beginning of the second hospitalization. The patient had anemia (Fig. 2A and B) and continued the previous treatment plan, including anti-CMV, anti-PJP, anti-COVID-19, anti-HIV, and added piperacillin/tazobactam solid on 30 June to antibacterial agents. However, the body temperature remained uncontrolled, with a maximum temperature of 39.8 °C (Fig. 2H). (2) Persistent fever and Pulmonary Progression (20230703–20230710): The doctor adjusted the patient's medication regimen by using TIENAM antibacterial, voriconazole antifungal, Ganciclovir + Foscaret anti-CMV, SMZ anti-PJP, Molnupiravir anti-COVID-19, and dexamethasone for anti-inflammatory (Fig. 1). The patient's infection indicators were high (Fig. 2C, D, 2E). On 10 July, the patient's lung CT showed diffuse infection in both lungs (Fig. 3C), with significant lung progression and blood gas analysis indicating type I respiratory failure (Fig. 2G). The persistent infection caused the patient to develop hyperlactatemia (Fig. 2F). The doctor empirically used drugs against *Mycobacterium* (isoniazid, linezolid, moxifloxacin, amikacin, and ethambutol) on July 10th (Fig. 1), but the anti-mycobacterium regime stopped after being transferred to the ICU. (3) ICU period (20230711–20230728): The patient underwent endotracheal intubation (ETI) as soon as he entered the ICU and continued until July 25 (Fig. 1). On July 17th, acid-fast bacilli were detected in the blood mycobacterium culture (BD BACTEC™ Myco/F Lytic Culture Vials, BD Bactec FX400) which was sampled on July 2nd (Table 2). Based on the patient's medical history, clinical manifestations, and imaging characteristics, a *non-tuberculosis mycobacterium* (NTM) infection was first considered. After consultation and discussion, it was decided to add anti-NTM drug regimens and cover tuberculosis treatment, with specific regimens including ethambutol, azithromycin, rifampicin, moxifloxacin, and amikacin. Considering the patient's lung CT (Fig. 3D) and poor oxygen index (Fig. 2G), extracorporeal membrane oxygenation (ECMO) was performed from July 17th to 24th to improve the patient's respiratory function. After a period of anti-NTM treatment, his pulmonary inflammation (Fig. 2C,

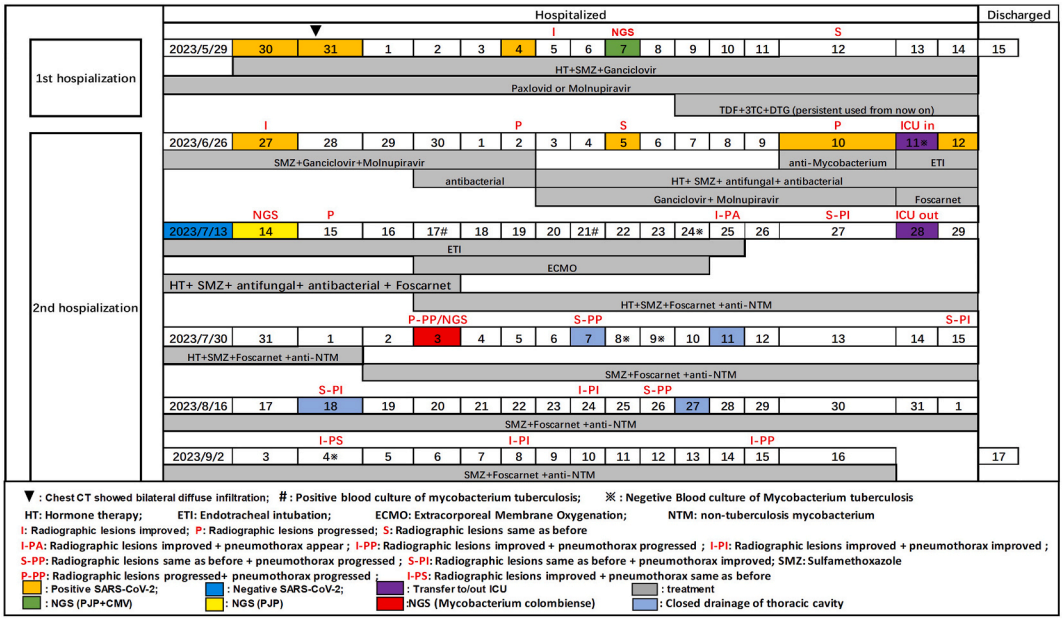


**Fig. 1.** Diagram of important events during the patient's two hospitalizations.

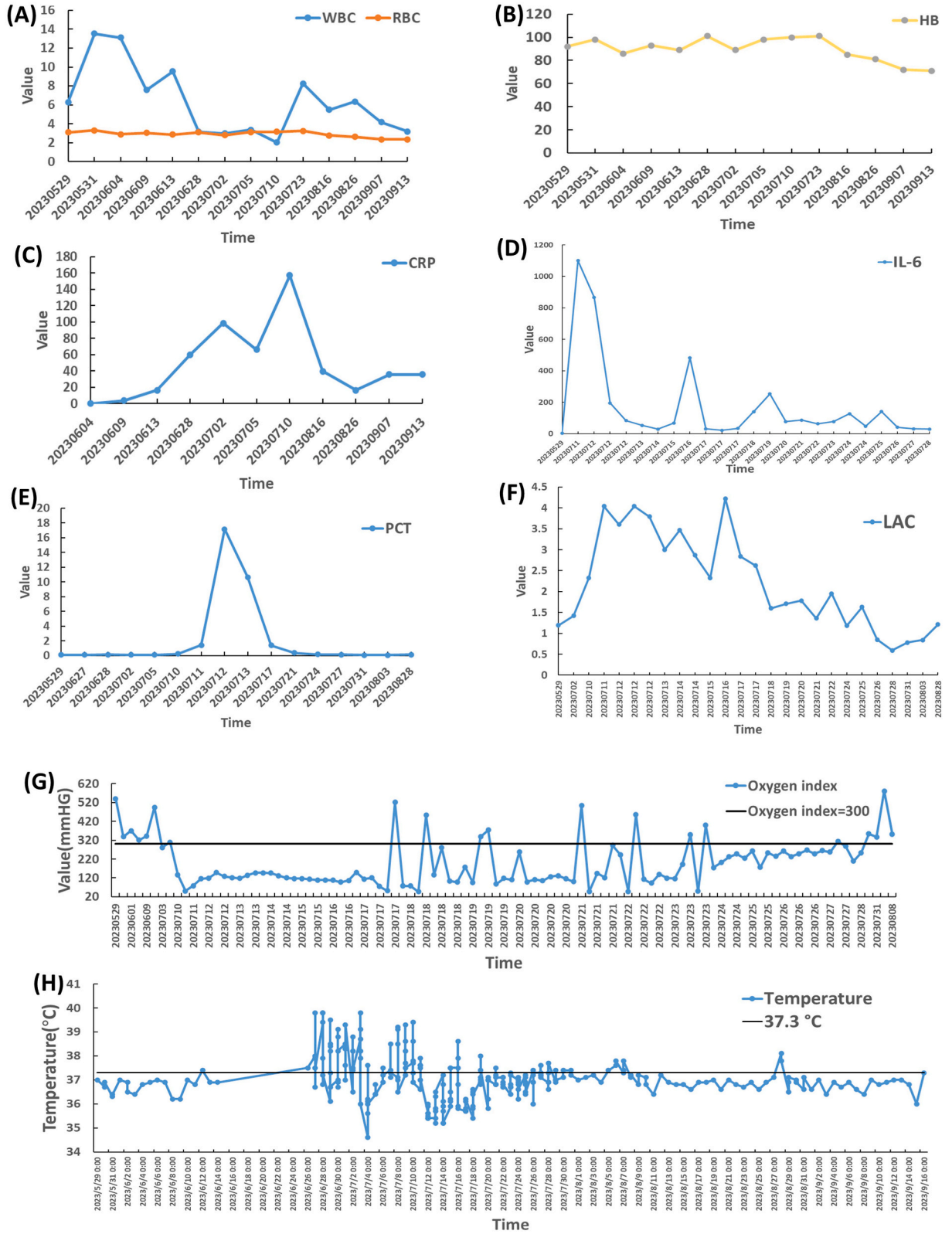


**Fig. 2.** Physical indicators of patients. (A) White Blood Cell (WBC) + Red Blood Cell (RBC); (B) Hemoglobin (HB); (C) C-reactive protein (CRP); (D) Interleukin-6 (IL-6); (E) Procalcitonin (PCT); (F) Lactic acid (LAC); (G) Oxygen index; (H) Body temperature.

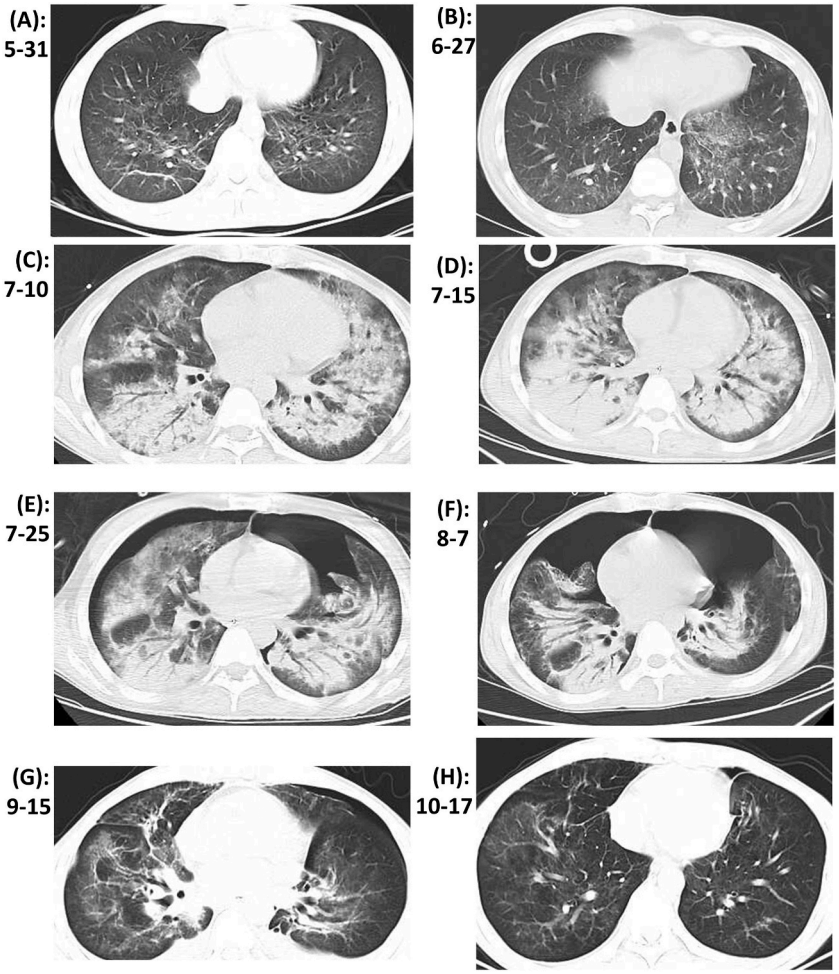


**Fig. 3.** Chest CT images. (A) 20230531 CT: the first chest CT in our hospital; (B) 20230627 CT: the first chest CT in the second hospitalization; (C) 20230710 CT: Progression of pulmonary inflammation; (D) 20230715 CT: Progression of pulmonary inflammation; (E) 20230725 CT: Radiographic lesions improved and pneumothorax appear; (F) 20230807 CT: obvious pneumothorax; (G) 20230915 CT: Radiographic lesions improved and pneumothorax improved; (H) 202301017 CT: one month follow-up, and the Radiographic lesions improved and pneumothorax improved.

**Table 2**
Results of bronchoalveolar lavage fluid (BALF) and blood culture at different periods.

| Sample time | Report time | Specimen type | Result |
|---|---|---|---|
| 20230529 | 20230711 | Blood culture | Negative |
| 20230606 | 20230607 | BALF NGS | CMV, PCP detected |
| 20230627 | 20230808 | Blood culture | Negative |
| 20230628 | 20230809 | Blood culture | Negative |
| 20230702 | 20230717 | Blood culture | Acid-fast bacilli |
| 20230710 | 20230721 | Blood culture | Acid-fast bacilli |
| 20230712 | 20230714 | BALF NGS | PCP detected |
| 20230724 | 20230904 | Blood culture | Negative |
| 20230731 | 20230803 | Blood culture specimen of 20230710 | M. Colombiense detected |
| 20230828 | 20231019 | Blood culture | Negative |
| 20231016 | 20231127 | Blood culture | Negative |

D, 2E) improved, body temperature (Fig. 2H) returned to normal, lung CT showed improvement (Fig. 3E), and he was transferred out of the ICU on July 28th. (4) Pneumothorax period (20230729–20230917): The patient continued to receive anti-CMV, anti-PJP, anti-HIV, and anti-NTM regimens. Pathogens targeted next generation sequencing (tNGS) using the MGISEQ-200 was used to identify the pathogens of the blood mycobacterium culture of 20230710 (Table 2), and the results showed that the sequence reads of *M. Colombiense* were 3135715.

The tNGS process was performed as below: 1): Extracting DNA from blood culture samples using TlANamp Micro DNA Kit (DP316, Tiangen Biotechnology, China); 2): Evaluating the concentration and purity of extracted genomic DNA using Qubit and Nanodrop techniques, and the concentration of starting DNA was 5.1ng/ul; 3) Qualified DNA library was prepared using One Shot DNA Library Prep Kit (PDM602, Nanjing Practice Medicine Diagnosis. Co., Ltd., China), and library DNA was 9.88ng/ul, and quality control was evaluated using an Agilent 2100 biological fragment analyzer; 4) DNA nanospheres were prepared and loaded onto sequencing chips for high-throughput sequencing using the MGISEQ-200 gene sequencer, and the pathogens panel was shown in Supplementary Table 2; 5) Bioinformatics analysis was performed on the raw data to obtain sequence alignment results.

The inflammatory absorption of the lung improved, but lung consolidation and pneumothorax also occurred (Fig. 3F). The patient underwent bilateral closed drainage of thoracic activity on 7 August (Fig. 1) but repeated pneumothorax and liquid pneumothorax occurred in the following period. The CT showed improvement on 15 September (Fig. 3G), and the patient was discharged from the hospital on 17 September. The patient's lung CT follow-up manifestations improved significantly (Fig. 3H) one month later after discharge. The patient returned to the hospital for follow-up every other month since the end of the second hospitalization. The latest inspection results were: CD4 140 cells/ul, HIV viral load <60 copies/ml, CMV viral load <500 copies/ml, and BMI 19.00 (Table 1).

## 3. Discussion

We present an AIDS patient coinfected with CMV, PJP, COVID-19, and M. Colombiense, who finally recovered from the above diseases. In the first hospitalization, the patient was diagnosed with CMV, PJP, HIV, and COVID-19 infection, and the corresponding treatment worked well. However, in the second hospitalization, the diagnosis of M. Colombiense was hard because the NGS, acid-fast bacilli, and culture of vomit, sputum, and BALF were all negative, but we detected acid-fast bacilli in blood mycobacterium culture and then used NGS to identify the specific pathogens. Previous case reports found M. Colombiense from the lymph node [11,17], sputum [13,14], pus sample [19], and BALF [13,16], but we found M. Colombiense in the blood. The same situation was also found in Mycobacterium monacense [20], which was isolated from the blood culture and the BALF NGS was negative. The possible explanation for the negative M. Colombiense in BALF NGS but positive in the blood culture can be as follows. First, the doctor empirically used anti-mycobacterium drugs on 10 July, but the BALF was taken on 12 July. Therefore, the drugs can affect M. Colombiense in the BALF. Second, the thick cell wall of mycobacterium made it difficult to destroy and extract nucleic acids in NGS tests. For HIV patients with pulmonary infections, we recommend that they not only undergo rapid and accurate NGS testing but also improve traditional microbial culture, especially for patients infected with NTM, who often can only be diagnosed through culture.

During the second hospitalization, a paradoxical IRIS was first diagnosed but was rejected by the patient's worsening condition. However, the finding of *M. Colombiense* and its anti-NTM treatment proved the patient has an "unmasking" IRIS. Previous studies [21–23] demonstrated baseline predictors, such as low CD4 count, low BMI, high HIV viral load, and low hemoglobin significantly increased the risk of developing IRIS. The patient had a baseline $CD4^{+}$ count of 95 copies/ul, BMI of 16.85, HIV viral load of 1.44*10^5 copies/ml, and anemia, which helped him develop the IRIS.

Most of the severe pneumonia patients infected with M. Colombiense died [13–15]. Fortunately, ECMO was performed when the patient had poor lung function, and the patient was finally rescued. ECMO [24,25] has been used as rescue therapy in appropriate COVID-19 and has saved many lives during the pandemic. After being transferred to the ICU, the traditional ETI method did not improve the patient's lung function and the oxygenation index still did not improve (Fig. 2G). However, ECMO was used on 17 July and the patient's oxygenation index improved significantly. Using the appropriate anti-NTM treatment, the patient's pulmonary inflammation improved, his body temperature returned to normal, and he was out of danger.

There were some limitations in this study. First, the patient was clinically considered to have a pulmonary infection at the second admission. Although pathogen testing was performed on sputum, BALF, and vomit, NGS testing was not performed on the blood. 2. During the hospitalization in the ICU, the patient's blood culture sample showed positive acid-fast bacilli and was not immediately tested for NGS. When the patient's condition was critical, empirical medication was used.

In conclusion, we report a severe pneumonia AIDS patient coinfected with CMV, PJP, COVID-19, and M. Colombiense who finally recovered from the disease. NTM infection is common in HIV patients, but BALF NGS cannot identify NTM in our report. Traditional blood culture was useful in detecting acid-fast bacilli in our study and detecting the pathogens with NGS. Combining traditional microbial culture and emerging rapid NGS methods is more conducive to clinical diagnosis and treatment.

## Patient consent statement

The study protocol followed the ethical standards of the institutional research committee and the ethics guidelines of the 1975 Declaration of Helsinki. Written informed consent was obtained from the patient for the publication of all images, clinical data, and other data included in the main manuscript. This study was approved by the institutional ethics review committee of Hangzhou Xixi Hospital.

## Data availability

Data will be made available on request.

## CRediT authorship contribution statement

**Zhongbao Zuo:** Writing – original draft, Formal analysis. **Rongrong Zheng:** Validation, Formal analysis. **Feng Li:** Visualization, Data curation. **Aifang Xu:** Supervision. **Jinchuan Shi:** Writing – review & editing, Supervision.

## Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e31729.

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