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Association between Clinical Characteristics and Microbiota in Bronchiectasis Patients Based on Metagenomic Next-Generation Sequencing Technology

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Abstract

This study aimed to investigate the disparities between metagenomic next-generation sequencing (mNGS) and conventional culture results in patients with bronchiectasis. Additionally, we sought to investigate the correlation between the clinical characteristics of patients and their microbiome profiles. The overarching goal was to enhance the effective management and treatment of bronchiectasis patients, providing a theoretical foundation for healthcare professionals. A retrospective survey was conducted on 67 bronchiectasis patients admitted to The First Hospital of Jiaxing from October 2019 to March 2023. Clinical baseline information, inflammatory indicators, and pathogen detection reports, including mNGS, conventional blood culture, bronchoalveolar lavage fluid (BALF) culture, and sputum culture results, were collected. By comparing the results of mNGS and conventional culture, the differences in pathogen detection rate and pathogen types were explored, and the diagnostic performance of mNGS compared to conventional culture was evaluated. Based on the various pathogens detected by mNGS, the association between clinical characteristics of bronchiectasis patients and mNGS microbiota results was analyzed. The number and types of pathogens detected by mNGS were significantly larger than those detected by conventional culture. The diagnostic efficacy of mNGS was significantly superior to conventional culture for all types of pathogens, particularly in viral detection ($p < 0.01$). Regarding pathogen detection rate, the bacteria with the highest detection rate were *Pseudomonas aeruginosa* (17/58) and *Haemophilus influenzae* (11/58); the fungus with the highest detection rate was *Aspergillus fumigatus* (10/21), and the virus with the highest detection rate was human herpes virus 4 (4/11). Differences were observed between the positive and negative groups for *P. aeruginosa* in terms of common scoring systems for bronchiectasis and whether the main symptom of bronchiectasis manifested as thick sputum ($p < 0.05$). Significant distinctions were also noted between the positive and negative groups for *A. fumigatus* regarding Reiff score, neutrophil percentage, bronchiectasis etiology, and alterations in treatment plans following mNGS results reporting ($p < 0.05$). Notably, 70% of patients with positive *A. fumigatus* infection opted to change their treatment plans. The correlation study between clinical characteristics of bronchiectasis patients and mNGS microbiological results revealed that bacteria, such as *P. aeruginosa*, and fungi, such as *A. fumigatus*, were associated with specific clinical features of patients. This underscored the significance of mNGS in guiding personalized treatment approaches. mNGS could identify multiple pathogens in different types of bronchiectasis samples and was a rapid and effective diagnostic tool for pathogen identification. Its use was recommended for diagnosing the causes of infections in bronchiectasis patients.

K e y w o r d s: bronchiectasis, mNGS technology, microbiological culture, infection

Introduction

Bronchiectasis is a common chronic lung disease characterized by irreversible dilation of the bronchi with a diameter greater than 2 mm caused by factors such as infection, physical and chemical factors, immune dysfunction, or genetic factors (Gao et al. 2021). Patients often experience acute exacerbations, presenting with symptoms such as fever, sputum production, purulent sputum, and progressive respiratory distress (Evans and Greenstone 2003; Amati et al. 2019). A study on bronchiectasis among adults in urban areas of mainland

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China in 2022 showed that the prevalence had increased by 2.31 times, from 75.48 per 100,000 in 2013 to 174.45 per 100,000 in 2017 (Feng et al. 2022). Studies have shown that the incidence and prevalence of bronchiectasis in the UK are increasing annually and are significantly associated with increased mortality rates (Quint et al. 2016), while the overall prevalence of bronchiectasis in Germany increases by approximately 10% per year (Ringshausen et al. 2019). Bronchiectasis poses an increasing burden on healthcare systems worldwide. Given that bronchiectasis is characterized by complex interactions between chronic infection, inflammation, and impaired mucociliary clearance leading to structural changes in the lungs, infections often result in acute exacerbations and worsening of the disease (Flume et al. 2018; Martinez-García 2018), necessitating early identification of infective pathogens and proactive use of antibiotics to control the condition (Bilton et al. 2006; Byun et al. 2017). Conventional pathogen detection methods include microbial culture, microscopy, histopathology, and polymerase chain reaction (PCR). However, microbial culture and microscopy have limitations in terms of detection rate and available targets, thereby failing to meet the needs of clinical practitioners (Zhang et al. 2020). Additionally, histopathological analysis is time-consuming and lacks pathogen specificity (Guarner and Brandt 2011), while PCR assays require the design of specific primers or probes for microbial pathogens, limiting the ability to detect pathogens (Maartens et al. 2020). When microorganisms cannot be promptly identified, physicians often rely on empirical antibiotic use, which is often ineffective and may exacerbate reinfection and accelerate the emergence of antibiotic resistance and multidrug-resistant pathogens (Ruppé et al. 2016). Therefore, it is necessary to establish a rapid and accurate method for detecting multiple pathogenic microorganisms in bronchiectasis patients.

Metagenomic next-generation sequencing (mNGS) technology provides a powerful solution to these clinical diagnostic challenges. The unbiased molecular technique of mNGS can simultaneously identify bacteria, viruses, fungi, parasites, and other microorganisms in clinical specimens by detecting their entire DNA and/or RNA content. It offers advantages such as high precision, high sensitivity, and short detection time (Chiu and Miller 2019; Chen et al. 2021; Li et al. 2023), making it beneficial for identifying severe infections, mixed infections, and uncommon or novel pathogen infections in immunocompromised patients (Chiu and Miller 2019). Previous studies have applied mNGS in the microbial diagnosis of infectious diseases such as sepsis and acute respiratory distress syndrome (Zhang et al. 2020; Ren et al. 2021). However, there is currently no research investigating the use of mNGS in the microbial diagnosis of bronchiectasis.

Additionally, in clinical practice, there is a lack of exploration into the correlation between clinical characteristics and microbiome. This could assist physicians in rapidly determining the cause of a patient's condition and consequently prescribing accurate medications. Therefore, this study aimed to analyze and compare the results of patients' microbiomes obtained through mNGS and conventional culture. The goal was to provide a more comprehensive, rapid, and accurate method for pathogen detection, thoroughly revealing the pathogen spectrum in patients with bronchiectasis. This, in turn, aided in formulating targeted therapeutic strategies, guiding improvements in patient prognosis care, and ultimately enhancing the quality of life for patients.

Experimental

Materials and Methods

Study population and sample collection. A retrospective analysis was conducted on 67 patients with bronchiectasis admitted to The First Hospital of Jiaxing from October 2019 to March 2023. The inclusion and exclusion criteria for patients were based on the expert consensus on the diagnosis and treatment of adult bronchiectasis in China published in 2021 (Bronchiectasis Expert Consensus Writing and Pulmonary Infection Assembly of Chinese Thoracic Society 2021). The inclusion criteria were as follows: 1) Clinical history consistent with bronchiectasis, confirmed by results of high-resolution computed tomography (HRCT) imaging of the chest, including both direct and indirect signs: a) bronchial diameter/adjacent pulmonary artery diameter > 1, b) gradual tapering of the bronchi from the center to the periphery, c) visible bronchi within 1 cm of the peripheral pleura or close to the range of the mediastinal pleura. Indirect signs included a) thickening of the bronchial wall, b) mucus plugging, and c) "mosaic" or "air trapping" signs detected on expiratory CT. 2) Age 18 and above. 3) Underwent mNGS pathogen detection, and the results were complete. The exclusion criteria were: 1) traction bronchiectasis associated with interstitial lung disease or other pulmonary diseases, 2) patients who were unable or unwilling to provide informed consent. Informed consent forms were signed by all patients or their legal guardians voluntarily participating in the study.

Blood, bronchoalveolar lavage fluid (BALF), and sputum specimens were collected from the patients after admission for routine microbial culture and mNGS testing.

Data collection. A retrospective survey was conducted to collect baseline information on patients, including age, gender, length of hospital stay, body mass

index (BMI), number of acute exacerbations of bronchiectasis within the past 12 months, number of hospitalizations due to acute exacerbations within the past 12 months, Bronchiectasis Severity Index (BSI) score, E-FACED score, BACI score, Reiff score, systolic and diastolic blood pressure, respiratory rate, pulse rate, etiology of bronchiectasis, main symptoms of bronchiectasis, smoking history, and other baseline information. Inflammatory indicators such as white blood cell count, neutrophil ratio, and neutrophil count were collected. The sequencing results of mNGS, routine blood culture, BALF culture, sputum culture, and information on the period of mNGS reports and other pathogen detections were also collected. The main clinical outcome measures in this study were whether the treatment plan was modified based on the mNGS results and the final clinical outcome of the patients.

mNGS. Specimens for mNGS sequencing were sent to the BGI Genomics (China) and Qingdao Ruiyi Precision Medical Laboratory Co., Ltd. (China) for pathogen detection. After receiving the specimens, the laboratory followed standard operating procedures for sample processing, nucleic acid extraction, DNA library preparation, high-throughput sequencing, bioinformatics analysis, and interpretation of mNGS data.

The main steps are as follows:

1) construction of DNA libraries through DNA fragmentation, end repair, adapter ligation, and PCR amplification; 2) quality control of DNA libraries using Agilent 2100 (Agilent Technologies, USA) (Davies et al. 2016); 3) sequencing of qualified libraries on the BGISEQ-50 platform (BGI, China) (Jeon et al. 2014); 4) raw data undergo quality control using fastp v0.19.4 (Chen et al. 2018) (sequences with lengths < 50 bp are filtered, bases with Phred quality < 20 are filtered, sequences with more than 40% of bases not meeting quality standards are filtered, sequences with more than three "N" bases are filtered). The post-sequencing data is stored on the hard drive in fastq format after conversion; 5) calculation of high-quality sequencing data by subtracting sequences mapped to the human reference genome (hg19) using Burrows-Wheeler Alignment (Li and Durbin 2009); 6) quality assessment of sequences using fastqc v0.11.5 (de Sena Brandine and Smith 2019); 7) remaining data after removing low-complexity reads are classified by simultaneously aligning to four microbial genome databases (including viruses, bacteria, fungi, and parasites). The reference databases for classification are downloaded from NCBI (ftp://ftp.ncbi.nlm.nih.gov/genomes), and Kraken 2 is used (Wood et al. 2019; He et al. 2022). In this study, a bacterium or virus was considered mNGS-positive if the number of reads for that microorganism was ten times higher than that of any other microorganism, and a fungus was considered mNGS-positive if its coverage was five times higher than that of another fungus (Yan et al. 2020; Yang et al. 2022).

Statistical analysis. All data in the study were statistically analyzed using IBM® SPSS® Statistics version 26.0 (IBM Corp., USA), and GraphPad version 8.0 (GraphPad Software, USA; www.graphpad.com) was used for data visualization. Descriptive analysis was performed using mean ± standard deviation (mean ± SD) for continuous variables and counts (n (%)) for categorical variables. Independent sample *t*-tests were used to compare normally distributed continuous variables, and Mann-Whitney *U*-tests were used to analyze non-normally distributed data. Pearson's chi-square or Fisher's exact test was used to analyze categorical variables. In this study, a two-sided p-value < 0.05 was considered statistically significant.

Results

Clinical characteristics of patients with bronchiectasis. A total of 67 bronchiectasis patients who underwent mNGS testing were included in this study. However, not all patients had complete clinical information. The specific baseline characteristics and clinical features are listed in Table I. Among the 67 patients, there were 37 female patients (55.2%) and 30 male patients (44.8%). Eight patients had a history of smoking (11.9%). After undergoing mNGS testing, 22 patients (32.8%) had their treatment plan modified, while 45 patients (67.2%) continued with the original treatment plan. Regarding the clinical outcome, 61 patients (91.0%) showed improvement in their condition, while six patients (9.0%) did not show improvement.

Comparison of mNGS and conventional cultures in diagnostic performance. Comparing the diagnostic performance of mNGS sequencing, blood culture, BALF culture, and sputum culture, it was found that mNGS detected a significantly larger number and variety of pathogens compared to conventional culture methods such as blood culture, BALF culture, and sputum culture ($p < 0.001$). Moreover, only mNGS could detect viral pathogens ($p < 0.001$), while conventional culture methods failed to detect viral pathogens. These results indicated that mNGS had significantly superior diagnostic performance for various pathogens compared to conventional cultures, particularly in viral detection (Table II, Fig. 1).

Detection results of pathogens. The four detection methods in this study identified a total of 52 pathogens, including 33 bacterial species, 13 fungal species, and six viral species. The specific pathogens are shown in Fig. 2. According to the pathogen detection results in Fig. 2, the most commonly detected bacterial pathogens were *Pseudomonas aeruginosa* (17/58) and *Haemophilus*

Table I
Summary of patient baseline characteristics.

Patient characteristic	Value
Days of hospitalization (days, n = 67)	8.64 ± 4.93
Age (years, n = 67)	61.72 ± 12.21
BMI (kg/m^2, n = 65)	20.60 ± 3.04
Number of acute exacerbations within 12 months (n = 67)	0.34 ± 0.54
Number of hospitalizations for acute exacerbations within 12 months (n = 67)	0.31 ± 0.53
BSI score (points, n = 67)	5.13 ± 3.19
E-FACED score (points, n = 67)	1.78 ± 1.57
BACI score (points, n = 67)	1.87 ± 2.81
Reiff score (points, n = 67)	2.84 ± 2.29
mNGS reporting period (days, n = 61)	1.61 ± 2.46
Systolic blood pressure (mmHg, n = 67)	130 ± 21.91
Diastolic blood pressure (mmHg, n = 67)	75.52 ± 11.92
Pulse (beats/min, n = 67)	86.61 ± 17.48
Respiration (beats/min, n = 67)	20.07 ± 2.72
White blood cell count (×10^9, n = 66)	7.28 ± 12.06
Neutrophils ratio (%, n = 66)	67.89 ± 10.16
Neutrophil count (×10^9, n = 66)	4.09 ± 2.07
Gender (n = 67)	
Male	30 (44.8%)
Female	37 (55.2%)
Etiology of bronchiectasis (n = 67)	
Bronchiectasis pulmonary aspergillosis (ABPA)	5 (7.5%)
Chronic obstructive pulmonary disease (COPD)	5 (7.5%)
Tuberculosis (TB)	5 (7.5%)
Infection	2 (3.0%)
Unknown cause	50 (74.6%)
Main symptoms of bronchiectasis (n = 67)	
Cough	51 (76.1%)
Thick sputum	25 (37.3%)
Hemoptysis	22 (32.8%)
Fever	7 (10.4%)
Other	20 (29.9%)
Smoking (n = 67)	8 (11.9%)
Whether to change treatment regimen based on mNGS results (n = 67)	
Yes	22 (32.8%)
No	45 (67.2%)
Disease outcome (n = 67)	
Improved	61 (91.0%)
Not improved	6 (9.0%)

Table II
Comparison of diagnostic efficacy of mNGS and conventional cultures (n = 67).

Group	mNGS	Blood culture	BALF culture	Sputum culture	*p*-value
All pathogens	66 (98.5%)	0	19 (28.4%)	18 (17.5%)	< 0.001
Bacteria	58 (86.6%)	0	10 (14.9%)	7 (10.4%)	< 0.001
Fungi	21 (31.3%)	0	9 (13.4%)	13 (19.4%)	< 0.001
Viruses	11 (16.4)	0	0	0	< 0.001

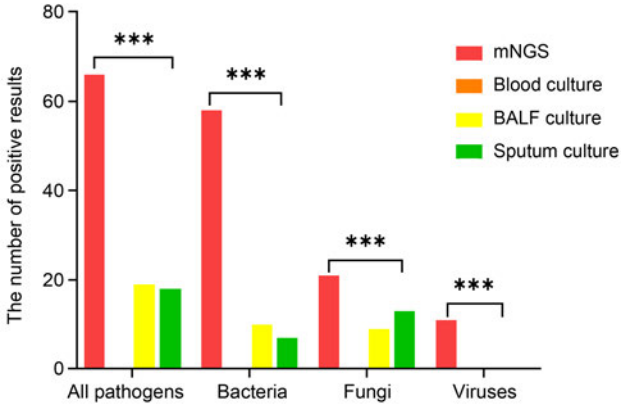


Fig. 1. Comparison of diagnostic performance between mNGS and conventional culture.
***$p < 0.001$

influenzae (11/58), the most commonly detected fungal pathogen was *Aspergillus fumigatus* (10/21), and the most commonly detected viral pathogen was human herpes virus 4 (4/11). Fig. 3 provides a visual representation of the specific number of pathogen species detected by the four different detection methods. It could be observed that mNGS sequencing detected 33 bacterial species, 10 fungal species, and six viral species, BALF culture detected four bacterial species and four fungal species, sputum culture detected three bacterial species and four fungal species, while blood culture did not detect any pathogens (Fig. 3).

Association between clinical characteristics of bronchiectasis patients and mNGS microbiological results. This study further investigated the relationship



Fig. 2. Microbial results of the four detection methods.

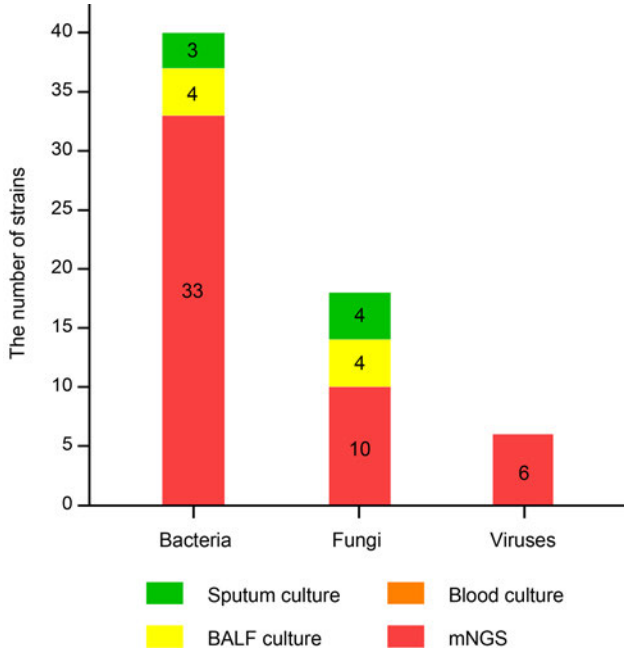


Fig. 3. Detection results of the four detection methods for different pathogens.

between the detection results of different pathogen types and the clinical characteristics of bronchiectasis patients. The relationship between bacterial detection results and patient clinical characteristics is explored in Table SI. The results showed no significant differences in clinical characteristics between the bacterial-positive and bacterial-negative groups ($p > 0.05$). The relationship between fungal detection results and patient clinical characteristics is examined in Table SII. The results revealed significant differences in the etiology of bronchiectasis, smoking status, and treatment plan modifications based on mNGS reports between the fungal-positive and fungal-negative groups ($p < 0.05$). The relationship between viral detection results and patient clinical characteristics is shown in Table SIII. The results showed a significant difference in the pulse rate per minute between the viral-positive and viral-negative groups ($p < 0.05$).

In subsequent research, based on the types of pathogens detected using the four detection methods, with a minimum detection count of ≥ 10, we selected the two most commonly detected bacterial pathogens *P. aeruginosa* (17/58) and *H. influenzae* (11/58); the most commonly detected fungal pathogen: *A. fumigatus* (10/21). We investigated the relationship between the specific pathogens detected by mNGS and patient clinical characteristics, as shown in Tables SIV, SV and SVI. The data results revealed significant differences in BSI score, E-FACED score, BACI score, Reiff score, and whether the main symptom of bronchiectasis was productive cough between the *P. aeruginosa*-positive and *P. aeruginosa*-negative groups ($p < 0.05$, Table SIV). The results in Table SV shows no significant differences in features between the *H. influenzae*-positive and *H. influenzae*-negative groups ($p > 0.05$). Table SIV demonstrates significant differences in Reiff score, neutrophil ratio, and bronchiectasis etiology between the *A. fumigatus*-positive and *A. fumigatus*-negative groups ($p < 0.05$). Additionally, a significant difference was observed in treatment plan modifications based on mNGS reports between the *A. fumigatus*-positive and *A. fumigatus*-negative groups ($p < 0.05$), with the majority of patients (70%) with *A. fumigatus* infection changing their original treatment plan.

Discussion

Numerous studies have demonstrated that worsening of bronchiectasis leads to increased airway and systemic inflammation, accompanied by progressive lung damage, decreased quality of life, accelerated decline in lung function, and increased mortality rates (Chalmers et al. 2012; Chalmers et al. 2014; Aliberti et al. 2016). This imposes a heavy burden on patients regarding medical expenses and prognosis. Acute exacerbations of bronchiectasis are often attributed to bacterial infections, and the Updated BTS Adult Bronchiectasis Guideline 2018 recommends antibiotic therapy to prevent or control acute exacerbations (Hill et al. 2019). Therefore, timely and effective identification of the microbial infection types in bronchiectasis patients and the use of targeted therapeutic drugs are crucial. This study, based on mNGS technology, investigated the association between clinical characteristics of bronchiectasis patients and the microbiome. The study results showed that the types and number of pathogens detected by mNGS were significantly higher than those detected by conventional culture methods. The diagnostic performance for various pathogens was significantly more robust with mNGS compared to conventional culture methods, particularly in viral detection, where it had unmatched advantages. The most detected bacterial pathogens among the bacterial types were *P. aeruginosa* and *H. influenzae*, while the highest-detected fungal pathogen was *A. fumigatus*.

This study compared the effectiveness of mNGS with traditional microbiological detection methods. The results showed that the overall positivity rate of mNGS (n = 66, 98.5%) was significantly higher than that of BALF culture (n = 19, 28.4%), sputum culture (n = 18, 17.5%), and blood culture (n = 0), indicating that only mNGS could detect viruses. This conclusion is similar to a previous study by Qi et al. (2019) on patients with related pneumonia, which found a much higher positivity rate with mNGS compared to conventional culture, and mNGS can to detect pathogens

that are difficult to detect with traditional methods. Cox et al. (2017) obtained similar results by using 16S rRNA. Furthermore, a study by Ren et al. (2021) investigating the diagnostic performance of mNGS in septic patients also demonstrated that mNGS can identify various pathogens in blood, BALF, and cerebrospinal fluid samples, showing a higher positivity rate compared to culture-based diagnostic methods (Ren et al. 2021). This is because mNGS is an unbiased and culture-independent method that employs high-throughput/parallel sequencing technology capable of simultaneously sequencing thousands of nucleic acid fragments. It analyzes nucleic acids in samples to detect and identify microbial DNA and/or RNA. The application of nanopore sequencing technology has significantly reduced the time from sample reception to final results, from 48 hours to 6 hours (Gu et al. 2019; Diao et al. 2022; Li et al. 2022).

Mainly, mNGS demonstrates pronounced advantages in detecting *Mycobacterium tuberculosis*, viruses, anaerobic bacteria, and fungi (Mei et al. 2023). Viruses require parasitism within host cells for survival and replication, making them difficult to culture using conventional methods. mNGS exhibits higher sensitivity and specificity than microbial culture, showing superior diagnostic performance over traditional methods (Qian et al. 2020).

Furthermore, compared to culture-dependent methods, mNGS results are less likely to be influenced by previous antibiotic exposure (Miao et al. 2018; Diao et al. 2022). Additionally, the results of this study also indicated that mNGS could be used to identify local infections. In this study, BALF and sputum samples were used for mNGS testing, and the results showed a high positivity rate, indicating the broad applicability of mNGS in pathogen detection, even in samples with relatively low positivity rates based on traditional culture-based diagnostic procedures (Ren et al. 2021). These findings again confirm the excellent diagnostic performance of mNGS in infectious diseases. The diagnostic method contributes to improving the clinical identification of infections of unknown origin.

In addition, the mNGS sequencing results of this study showed that *P. aeruginosa* and *H. influenzae* were the most common pathogens in patients with bronchiectasis, ranking first and second, respectively. This result is similar to a previous study on secreted mucins and airway bacterial colonization in 50 non-cystic fibrosis bronchiectasis patients from Europe (Sibila et al. 2015). However, according to the results reported by the US Bronchiectasis Research Registry, *P. aeruginosa* has a higher isolation rate (33%) in bronchiectasis patients, while *H. influenzae* is relatively uncommon (8%) (Aksamit et al. 2017). These discrepancies may be due to differences in patient sample selection, collection methods, and geographic variations in microbial characteristics. Furthermore, *P. aeruginosa* infection has been included as an important risk factor for disease prognosis and severity in various scoring systems used to assess the severity of bronchiectasis, such as the E-FACED score and BSI score (Martinez-Garcia et al. 2017; Gao et al. 2021). Most microbiological studies on bronchiectasis focus on bacteria, while fungi are generally considered incidental findings (Chandrasekaran et al. 2018). *Aspergillus* species are the most commonly isolated fungi in the sputum of bronchiectasis patients. *A. fumigatus* can act as a pathogen or an allergen, but its specific role in bronchiectasis is still unclear (Kosmidis and Denning 2015; Amati et al. 2019). However, due to the high specificity and sensitivity of mNGS technology (Wang et al. 2019), it is possible to detect colonization of non-pathogenic bacteria. Respiratory colonizers (such as *P. aeruginosa*, *H. influenzae*, and *A. fumigatus*) maintain a balance with the host by evading the immune system's clearance (Zhao et al. 2023). Also, bronchiectasis patients are susceptible to bacterial infections (Chalmers et al. 2018). However, the microbial composition detected by mNGS cannot determine the pathogenic state of the detected microorganisms. Therefore, even though this study observed a high detection rate of *P. aeruginosa*, *H. influenzae*, and *A. fumigatus*, it is difficult to determine the true pathogenic microorganism. This is one of the limitations affecting the clinical application of mNGS. The impact of colonizers on respiratory tract structure and function could be a future research direction to explore the specific etiology of bronchiectasis. Based on the results of this study, we believe that in bronchiectasis patients, it may be necessary to reduce the carriage of potential pathogens in the oropharynx, stomach, intestines, or respiratory tract by using non-absorbable antibiotics or inhaled antibiotics selectively, while enhancing the patient's immune system to resist pathogenic microorganisms.

In this study, clinicians adjusted the anti-infection treatment based on mNGS results, which assisted clinical diagnosis. The results showed that in the group with positive *A. fumigatus* results, 70% of patients had their empirical anti-infection therapy adjusted according to mNGS results, and the outcomes of all patients showed improvement (100%). This indicates a positive impact of mNGS testing on clinical drug guidance and patient prognosis. The overall data results of the association between clinical characteristics of bronchiectasis patients and mNGS microbial results in this study demonstrated that the microbial composition detected in bronchiectasis patients is mainly related to common scoring systems such as Reiff, BSI, E-FACED, BACI scores, and some common clinical characteristics (Kosmidis and Denning 2015; Amati et al. 2019; Ma et al. 2021; Huang et al. 2023). These characteristics are

evaluation indicators of bronchiectasis and microbial infection, but they do not guide the etiological diagnosis of bronchiectasis pathogens in clinical practice. This indirectly indicates the necessity and rationality of laboratory pathogen detection for diagnosing infectious causes. In general, testing facilities can provide mNGS reports within 24 hours, and in practical treatment, this does not significantly increase the waiting time for patients. On the contrary, it is even shorter than the time required for conventional culture. Therefore, when bronchiectasis patients are at risk of infection, NGS can supplement routine microbial detection methods to increase the detection rate, shorten the turnaround time, and assist in diagnosing the etiology of patients for targeted treatment. Certainly, we hope to establish closer collaboration between hospitals and enterprises in the future, allowing the seamless deployment of the testing platform within hospital settings to achieve real-time detection. This will help reduce patient waiting times, enabling healthcare professionals to administer personalized treatments and enhance medical efficiency promptly. Additionally, it contributes to minimizing sample wastage and reducing transportation costs, alleviating the overall burden on healthcare.

Although encouraging results were achieved, it must be acknowledged that this study had certain limitations. Firstly, there was no authoritative and unified guideline for interpreting mNGS reports, which may lead to subjective misjudgments. Secondly, the sample size in this study was relatively small, which may introduce bias in the data. For instance, in the part of the study that examined the association between clinical characteristics and mNGS microbial results in patients with bronchiectasis, there were significant differences between the samples of pathogen-positive and pathogen-negative groups. Additionally, this study did not investigate the specific subtypes of single pathogen infection/mixed infection, nor did it consider the possibility of dominant bacterial populations within mixed infection pathogens, which may introduce bias in the results. In conclusion, the results of this study demonstrated the important significance of mNGS in the diagnosis, treatment, and prognosis of bronchiectasis patients. It is clinically recommended to use mNGS as a supplementary tool to routine microbial testing to improve the detection rate and assist in treatment decisions.

Availability of data and material

The data and materials in the current study are available from the corresponding author on reasonable request.

Author contributions

DF S, XD L, H Z, CY F and J F conceived of the study, and participated in its design and interpretation and helped to draft the manuscript. JQ Z, LF C and Y Y participated in the design and interpretation of the data and drafting/revising the manuscript. N L and XL M performed the statistical analysis and revised the manuscript critically. All the authors read and approved the final manuscript.

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Conflict of interest

The authors do not report any financial or personal connections with other persons or organizations, which might negatively affect the contents of this publication and/or claim authorship rights to this publication.

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