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Isolate distribution and antifungal susceptibility of *Saccharomyces cerevisiae* in the national regional medical center of Southwest China for women and children during 2018–2023

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Abstract

Background *Saccharomyces cerevisiae* has been considered a harmless yeast, but in recent years, increasing evidence has shown that it can cause disease in humans, especially invasive infections in infants/children and vulvovaginal infections in women. This study aimed to investigate the clinical information and antifungal susceptibility of clinical cases with *S. cerevisiae* and establish a foundation for the prevention and treatment of fungal infections.

Methods This study was conducted from May 2018 to May 2023 at a national regional medical center in Southwest China for women and children. The demographic and clinical characteristics of patients isolated with *S. cerevisiae* were collected and analyzed. All the isolates were cultured on Sabouraud medium plates and identified by MALDI-TOF MS. The antifungal susceptibility of *S. cerevisiae* to 10 agents (amphotericin B, fluconazole, itraconazole, voriconazole, micafungin, caspofungin, terbinafine and 5-flucytosine) was determined via the microdilution broth method to determine the minimum inhibitory concentrations (MICs).

Results A total of 75 cases of *S. cerevisiae* isolated from patients with vulvovaginal candidiasis (VVC, 44 cases), pneumonia (13 cases), or diarrhea (18 cases) were included after data review. The MICs of voriconazole and flucytosine for *S. cerevisiae* isolated from different body sites differed, with higher resistance in intestinal isolates. In this study, *S. cerevisiae* caused VVC, but there was no clear evidence that it was involved in pneumonia or diarrhea. Compared with those of *Candida albicans*, the primary pathogen of VVC, the MICs of fluconazole (11.96 ± 5.78 μg/mL vs. 67.64 ± 16.62 μg/mL, $p = 0.002$), itraconazole (0.77 ± 0.19 μg/mL vs. 2.31 ± 0.53 μg/mL, $p = 0.008$), voriconazole (0.22 ± 0.09 μg/mL vs. 5.02 ± 1.09 μg/mL, $p < 0.001$), and terbinafine (10.41 ± 0.84 μg/mL vs. 14.93 ± 4.77 μg/mL, $p < 0.001$) for *S. cerevisiae* (isolated from the genital tract) were significantly lower, while those of micafungin

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(0.14 ± 0.01 μg/mL vs. 0.06 ± 0.01 μg/mL, $p < 0.001$) and caspofungin (0.27 ± 0.04 μg/mL vs. 0.06 ± 0.01 μg/mL, $p < 0.001$) were significantly greater.

Conclusion Azoles remain the recommended regimen for *S. cerevisiae*-related VVC, and the use of amphotericin B vaginal effervescent tablets could be considered for the treatment of azole-resistant isolates. The antifungal susceptibility of *S. cerevisiae* varies according to the isolated source, and the pathogenicity trend of *S. cerevisiae* should be studied.

Highlights

- This is the first clinical *Saccharomyces cerevisiae* study conducted in East Asia and included 75 cases.
- *S. cerevisiae* can cause vulvovaginal candidiasis (VVC), but the difficulty of its treatment is no greater than that of *Candida albicans*.
- Azoles are still the recommended regimen for treating *S. cerevisiae* vulvovaginitis, and resistant isolates are usually susceptible to amphotericin B, for which vaginal effervescent tablets can be used.
- The antifungal susceptibility of clinical *S. cerevisiae* isolates may vary among regions, populations, and isolation sites, and attention should be given to the increasing trend of detection in medical institutions.

Keywords *Saccharomyces cerevisiae*, Vulvovaginal candidiasis, VVC, Antifungal susceptibility, Infection, Rare pathogenic fungi

Introduction

Saccharomyces cerevisiae (*S. cerevisiae*) is a species of yeast that is widely found in nature and the human environment, with cell shapes ranging from round to ovoid and 5–10 μm in diameter. It was originally isolated from the skin of grapes and has been widely used in various industrial endeavors, such as winemaking, baking, and brewing, since the 19th century [1, 2]. More recently, this species has also been used in bioindustries such as livestock feed, biotherapeutics (probiotics), bioremediation, and biofuels [3].

Although *S. cerevisiae* has long been considered a harmless industrial strain, there have been an increasing number of reports of *S. cerevisiae* causing disease in recent years [4]. As an opportunistic pathogenic fungus, *S. cerevisiae* has been shown to cause fungemia [5], fungal pneumonia [6], and vulvovaginal candidiasis (VVC) [7] in humans under certain conditions. It can also be detected at multiple sites in the human body, such as the oral cavity, bile duct, urine, and intestines [8]. Moreover, clinical studies have reported that even immunocompetent patients may suffer from *S. cerevisiae* infection [9].

Infants/children, women (VVC), elderly people, and immunosuppressed patients are the individuals most commonly infected with *S. cerevisiae* [4]. Children's immune systems are not fully developed, and their immune clearance capabilities against exogenous organisms are insufficient, making them susceptible to invasive infections. The female vaginal environment is acidic (pH < 4.5), and the vaginal epithelium is rich in glycogen, which is suitable for the growth of yeast-like fungi, including *S. cerevisiae*. These fungi, together with other colonizing bacteria, constitute the normal vaginal microecology system to resist the invasion of other pathogenic microorganisms. However, in some cases, yeast will transform into hyphae, grow pseudohyphae, invade the vaginal epithelium, and cause vulvar rash, itching, burning pain, pain during sexual intercourse, and urinary pain, resulting in VVC. When a VVC patient experiences symptoms three or more times within a year, she can be diagnosed with recurrent VVC (RVVC) [10].

In this study, we retrospectively investigated the clinical isolates of *S. cerevisiae* from a women's and children's specialized medical center (Southwest China) over the past five years, analyzed the clinical data, and performed antifungal susceptibility tests (AFSTs). This work aimed to enhance our understanding of *S. cerevisiae* isolated from humans at medical institutions, explore the susceptibility of isolates from different sites in the human body to various antifungal agents, and discuss treatment strategies for VVC patients. This study not only deepens the understanding of the clinical relevance of *S. cerevisiae* but also establishes a foundation for more targeted prevention and treatment strategies in the field of fungal infections.

Materials and methods

Patients

This is a retrospective study which conducted from May 2018 to May 2023 at West China Second University Hospital/National Regional Medical Center (Southwest China), which is one of the largest specialized hospitals for women and children in China.

First, we enrolled all patients with fungal culture identification of *S. cerevisiae* reported by clinical laboratories during this period (mixed flora were excluded from the experiment according to the CAP and ISO 15189 guidelines).

Second, we excluded patients with incomplete clinical data, repeated detection samples, and external quality control samples.

Third, the remaining patients were divided into 3 groups according to the following eligibility criteria: (1) Group VVC: patients were diagnosed with VVC according to the International Classification of Diseases 10th Revision (ICD-10), with *S. cerevisiae* isolated from vaginal or cervical secretions as single and uniform growth on Sabouraud dextrose agar (SDA) plates. (2) Group pneumonia: patients were diagnosed with pneumonia according to the ICD-10, with *S. cerevisiae* isolated from alveolar lavage fluid (ALF) or sputum as single and uniform growth on SDA plates. (3) Group diarrhea: patients were diagnosed with diarrhea according to the ICD-10, with *S. cerevisiae* isolated from feces or intestinal mucosa as single and uniform growth on SDA plates.

Fourth, stratified sampling of VVC patients caused by *C. albicans* according to patient age and the time of consultation were enrolled as [4] Group VVC (control). The flowchart is shown in Fig. 1.

Isolation and identification of strains

The hospital's clinical laboratory has been accredited by the College of American Pathologists (CAP) and the ISO 15,189 accreditation standard and quality control of the entire sample process were always followed, including initial sampling and final reporting. Specimens were collected by specialized physicians or nurses following the Standard Operating Procedure (SOP) of ISO/TS 20658:2017-Medical Laboratories-Requirements for collection, transport, receipt, and handing of samples (blood, secretions, sputum, feces, and intestinal mucosa). The ALF samples were collected by flexible bronchoscopy according to the protocol of the Chinese expert consensus on pathogen detection in bronchoalveolar lavage for pulmonary infectious diseases [11].

Strains were isolated on Sabouraud dextrose agar (SDA) plates (Autobio, Zhengzhou, China) and incubated at 35 °C for 24–72 h in a 5% carbon dioxide (CO_2) environment. All the isolates were identified by matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF MS; Vitek MS system; BioMerieux, Rhône, France) following the Clinical and Laboratory Standards Institute (CLSI) guideline M58: Methods for the Identification of Cultured Microorganisms Using MALDI-TOF MS. Quality control analysis of the MALDI-TOF MS identification system was performed with *Candida glabrata* (ATCC MYA-2950), *Candida parapsilosis* (ATCC 22019), *Enterococcus faecalis* (ATCC 19433) and *Escherichia coli* (ATCC 8739) based on the operation manual. In accordance with the reporting specifications, after inoculation, incubation and identification, the sample showed single and uniform growth of *Saccharomyces cerevisiae* colonies on SDA plates.

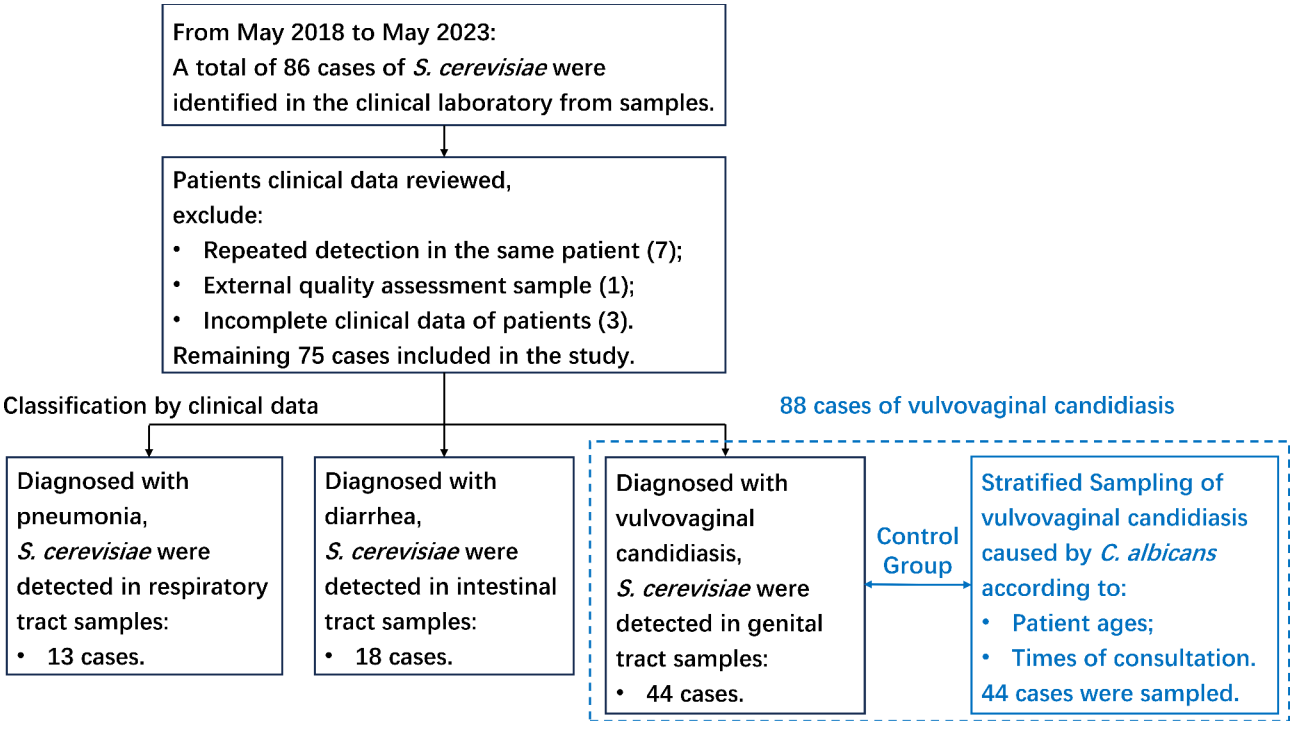

Fig. 1 Patient inclusion and exclusion process

Antifungal susceptibility tests

Antifungal susceptibility tests (AFSTs were performed using the broth turbidimetry method in AFST dishes (TDR YEAST-AST, Mindray, China). The antifungal agents used were amphotericin B, fluconazole, itraconazole, voriconazole, micafungin, caspofungin, terbinafine and 5-flucytosine. The operational processes were performed according to the manufacturer's instructions, and the results were interpreted by TDR-300B PLUS (Mindray, Hunan, China). Quality control analysis was performed with *Candida parapsilosis* (ATCC 22019).

Microscopic examination

Vaginal secretions and cervical secretions were smeared on glass slides with wet preparation and examined directly under a microscope to preliminarily and rapidly detect the presence of fungal spores. Feces were prepared as thin smears and Gram-stained for microscopic examination, and fecal dysbiosis was determined by the proportions of gram-positive bacilli, gram-negative bacilli, and gram-positive cocci. The reference interval and interpretation criteria were taken from the fecal smear atlas of intestinal flora (ISBN: 7-80157-147-9).

Table 1 Demographic and clinical characteristics of 75 *Saccharomyces cerevisiae* culture-positive patients

Characteristics	No. of patients	(%)
Sex		
Male	23	30.7
Female	52	69.3
Age (years)		
≤ 5	24	32.0
6–17	8	10.7
18–44	41	54.6
≥ 45	2	2.7
Median age (P25-P75)	*28 (1.3–31)*	
Sample type		
Vaginal secretions	41	54.7
Cervical secretions	3	4.0
Alveolar lavage fluid	11	14.6
Sputum	2	2.7
Feces	15	20.0
Intestinal mucosa	3	4.0
Diagnosis		
Vulvovaginal candidiasis	44	58.7
Pneumonia	13	17.3
Diarrhea	18	24.0
Prognosis		
Cured or improved	71	94.7
Recurred or quit therapy	4	5.3
Ineffective or exacerbated	0	0.0
Total	75	100.00

β-D-glucan test (G test) and galactomannan test (GM test)

Blood samples were collected from pneumonia patients and subjected to G tests and GM tests. The G test was performed using a fungus (1-3)-β-D-glucan test kit (chromogenic method, Gold Mountain River, Beijing, China) and a fungus dynamic detector (IGL-800, Genobio, Tianjin, China). A level of (1-3)-β-D-glucan ≥ 100 pg/mL was interpreted as positive according to the reference interval. The GM test was performed with an *Aspergillus* galactomannan test kit (chemiluminescence method, Genobio, Tianjin, China) and a chemiluminescence enzyme immunoassay analyzer (FACIS-I, Genobio, Tianjin, China). A level of galactomannan ≥ 0.45 μg/L was interpreted as positive according to the reference interval.

Detection of co-infectious pathogens

Sputum and ALF samples were inoculated on Columbia agar + 5% sheep blood plates (Autobio, Zhengzhou, China) and chocolate blood agar plates (Autobio, Zhengzhou, China) to isolate potentially pathogenic bacteria, and polymerase chain reaction (PCR) was performed to detect pathogenic viruses or *Mycoplasma*. The plates were incubated at 35 °C for 24–48 h in a 5% carbon dioxide (CO_2) environment, after which the suspected colonies were identified by MALDI-TOF MS (Vitek MS system; BioMerieux, Rhône, France).

Statistical analysis

Statistical Package for Social Sciences (SPSS) software for Windows was used to assess the statistical significance of the data (version 22.0; Chicago, IL, USA). In brief, one-way analysis of variance (ANOVA) and t-tests were used. The results of the one-way ANOVA were subjected to the least significant difference (LSD) method or Tamhane's method (depending on whether the variances were homogeneous) to determine whether differences among multiple groups were statistically significant. P values < 0.05 were considered statistically significant.

Results

Demographic and clinical characteristics

A total of 86 cases of *S. cerevisiae* infection were identified from May 2018 to May 2023, and 75 cases ultimately remained after the patients' clinical data were reviewed; these cases included pneumonia (13 cases), diarrhea (18 cases), and VVC (44 cases) (Fig. 1). Among the 75 patients, 52 patients were female (69.3%), with ages ranging from 0.09 to 48 years and a median age (P25-P75) of 28 (1.3–31) years. The samples included vaginal secretions, cervical secretions, ALF, sputum, feces, and intestinal mucosa, and 71 patients (94.7%) had a favorable prognosis (Table 1).

To obtain a comprehensive understanding of the VVC group, we conducted stratified sampling with VVC caused by *C. albicans* (44 cases), which is the primary pathogen that causes VVC in the study region, as a control group matched according to patient age and consultation time (Fig. 1). Among the 88 VVC patients (44 *S. cerevisiae* cases and 44 *C. albicans* cases), the ages ranged from 10 to 48 years, with median ages (P25-P75) of 30 (30–34) and 31 (28–34) years, respectively. Fungal spores were identified in the vaginal secretions of all the patients via direct smear microscopy, and the patients were treated with antifungal drugs; 10 patients (11.4%) developed RVVC (Table 2).

Table 2 Demographic and clinical characteristics of 88 vulvovaginal candidiasis patients

Characteristics	*Saccharomyces cerevisiae*		*Candida albicans*		*P*
	No. of patients	(%)	No. of patients	(%)	
Sex					NA
Male	0	0.0	0	0.0	
Female	44	100.0	44	100.0	
Age (years)					
≤ 17	1	2.3	1	2.3	
18–24	3	6.8	1	2.3	
25–34	32	72.7	32	72.7	
35–44	6	13.6	8	18.2	
≥ 45	2	4.5	2	4.5	
Median age (P25-P75)	*30 (30–34)*		*31 (28–34)*		0.950
Sample type					1.000
Vaginal secretions	41	93.2	41	93.2	
Cervical secretions	3	6.8	3	6.8	
Fungal spores found in vaginal secretions via microscopy					NA
Yes	44	100.0	44	100.0	
No	0	0.0	0	0.0	
Antifungal drug use[a]					NA
Yes	44	100.0	44	100.0	
No	0	0.0	0	0.0	
Prognosis					**0.044**
Cured or improved	42	95.5	36	81.8	
Recurrence	2	4.5	8	18.2	
Ineffective	0	0.0	0	0.0	
Total	44	100.0	44	100.0	

Note A case–control study was performed to evaluate the biology of *Saccharomyces cerevisiae* strains causing vulvovaginal candidiasis. *Candida albicans* is the primary pathogen causing vulvovaginal candidiasis in the study region. P values were evaluated by the Mann-Whitney U test, chi-square test and Fisher's exact test according to the data type and *P* values < 0.05 were considered statistically significant. a. Antifungal drugs, including amphotericin B, clotrimazole, fluconazole, miconazole nitrate and itraconazole

Antifungal susceptibility of 75 cases of *S. Cerevisiae* infection

The minimum inhibitory concentration (MIC) of each antifungal agent was evaluated and analyzed since the Clinical and Laboratory Standards Institute (CLSI) guidelines do not have resistance breakpoints or epidemiological cutoff values (ECVs) for *S. cerevisiae*. The MICs of the antifungal agents were as follows: (1) Amphotericin B concentrations ranged from 0.008 to 8 μg/mL, with a mean concentration (±Std. error of the mean) of 0.39±0.11 μg/mL. (2) Fluconazole concentrations ranged from 0.008 to 256 μg/mL, with a mean concentration (Std. error of the mean) of 8.04±3.42 μg/mL. (3) Itraconazole concentrations ranged from 0.008 to 8 μg/mL, with a mean concentration (Std. error of the mean) of 0.69±0.12 μg/mL. (4) Voriconazole concentrations ranged from 0.008 to 4 μg/mL, with a mean concentration (Std. error of the mean) of 0.29±0.07 μg/mL. (5) Micafungin concentrations ranged from 0.008 to 0.5 μg/mL, with a mean concentration (Std. error of the mean) of 0.14±0.01 μg/mL. (6) Caspofungin concentrations ranged from 0.008 to 16 μg/mL, with a mean concentration (Std. error of the mean) of 0.42±0.21 μg/mL. (7) Terbinafine concentrations ranged from 0.008 to 16 μg/mL, with a mean concentration (Std. error of the mean) of 10.72±0.63 μg/mL. (8) The 5-Flucytosine concentrations ranged from 0.12 to 0.5 μg/mL, with a mean concentration (Std. error of the mean) of 0.14±0.01 μg/mL (Fig. 2).

Antifungal susceptibility in 88 VVC patients

After antifungal treatment, *S. cerevisiae* caused a lower rate of recurrent vulvovaginal candidiasis (RVVC) than did *C. albicans* (4.5% vs. 18.2%, chi-square = 4.06, $p = 0.044$; Table 2). Among the pathogenic fungi isolated from the reproductive tract, the MICs of fluconazole (11.96±5.78 μg/mL vs. 67.64±16.62 μg/mL, $p = 0.002$), itraconazole (0.77±0.19 μg/mL vs. 2.31±0.53 μg/mL, $p = 0.008$), voriconazole (0.22±0.09 μg/mL vs. 5.02±1.09 μg/mL, $p < 0.001$) and terbinafine (10.41±0.84 μg/mL vs. 14.93±4.77 μg/mL, $p < 0.001$) for *S. cerevisiae* were significantly lower than those for *C. albicans*, while the MICs of micafungin (0.14±0.01 μg/mL vs. 0.06±0.01 μg/mL, $p < 0.001$) and caspofungin (0.27±0.04 μg/mL vs. 0.06±0.01 μg/mL, $p < 0.001$) for *S. cerevisiae* were significantly greater than those for *C. albicans.* No significant difference was found for the MICs of amphotericin B (0.25±0.03 μg/mL vs. 0.27±0.03 μg/mL, $p = 0.592$) or 5-flucytosine (0.13±0.01 μg/mL vs. 6.27±5.81 μg/mL, $p = 0.293$) (Fig. 3).

Taking the resistance breakpoints and ECV for *C. albicans* from the CLSI guidelines as references, the resistance of *S. cerevisiae* to voriconazole was significantly lower than that of *C. albicans* (4.5% vs. 47.7%, $p < 0.001$), whereas there were no significant differences

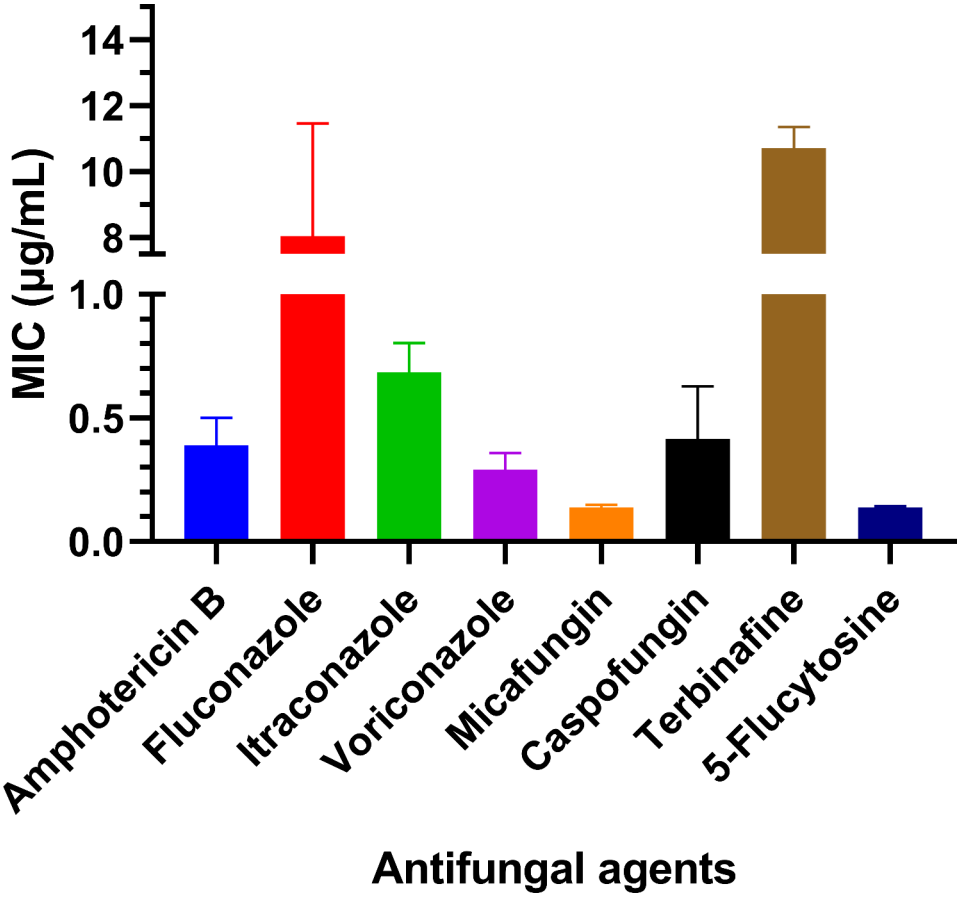

Fig. 2 Minimum inhibitory concentrations (MICs) of antifungal agents (mean ± SEM) against 75 *S. cerevisiae* isolates

in resistance to fluconazole (31.8% vs. 36.4%, $p=0.822$), micafungin (0.0% vs. 0.0%), caspofungin (9.1% vs. 0.0%, $p=0.116$) or amphotericin B (0.0% vs. 0.0%).

Antifungal susceptibility and clinical characteristics in pneumonia

A total of 13 cases of *S. cerevisiae* were isolated from pneumonia patients, including 10 boys (76.9%) and 3 girls (23.1%), with ages ranging from 0.18 to 12 years and a median age (P25-P75) of 0.50 (0.34–1.40) years. The samples included ALF and sputum (Table 3). *S. cerevisiae* was not considered to be the primary pathogen in any of the 13 cases; the primary pathogens included bacteria (*Staphylococcus aureus*, *Acinetobacter baumannii*, *Haemophilus influenzae*, etc.), viruses (SARS-CoV-2, human metapneumovirus, human bocavirus, etc.), *Mycoplasma* and *Aspergillus* (clinical immunology diagnosed). Four patients (30.8%) received prophylactic anti-invasive fungal infection treatment. Eleven patients (84.6%) had a favorable prognosis, and 2 families (15.4%) quit therapy during treatment (Table 3).

Among the 13 cases of *S. cerevisiae* isolated from the respiratory system, the mean MIC (±Std. error of the mean) of each antifungal agent was as follows: (1) amphotericin B: 0.39±0.14 μg/mL; (2) fluconazole: 4.89±0.78 μg/mL; (3) itraconazole: 0.53±0.09 μg/mL; (4) voriconazole: 0.09±0.01 μg/mL; (5) micafungin: 0.14±0.03 μg/mL; (6) caspofungin: 0.08±0.02 μg/mL; (7) terbinafine: 10.54±1.56 μg/mL; and (8) flucytosine: 0.13±0.01 μg/mL (Fig. 4).

Antifungal susceptibility and clinical characteristics in diarrhea

A total of 18 cases of *S. cerevisiae* were isolated from diarrhea patients, including 13 boys (72.2%) and 5 girls (27.8%), with ages ranging from 0.10 to 14 years and a median age (P25-P75) of 1.17 (0.23–7.25) years. The samples included feces and intestinal mucosa (Table 4). Although half of the patients had fecal flora dysbiosis, there was no clear clinical evidence that *S. cerevisiae* was the primary pathogen, and all patients had a favorable prognosis.

The antifungal spectrum of *S. cerevisiae* isolated from the digestive system was heterogeneous with those isolated from the reproductive system and the respiratory system. The MIC of voriconazole against isolates from the digestive system was significantly greater than that against isolates from the respiratory system

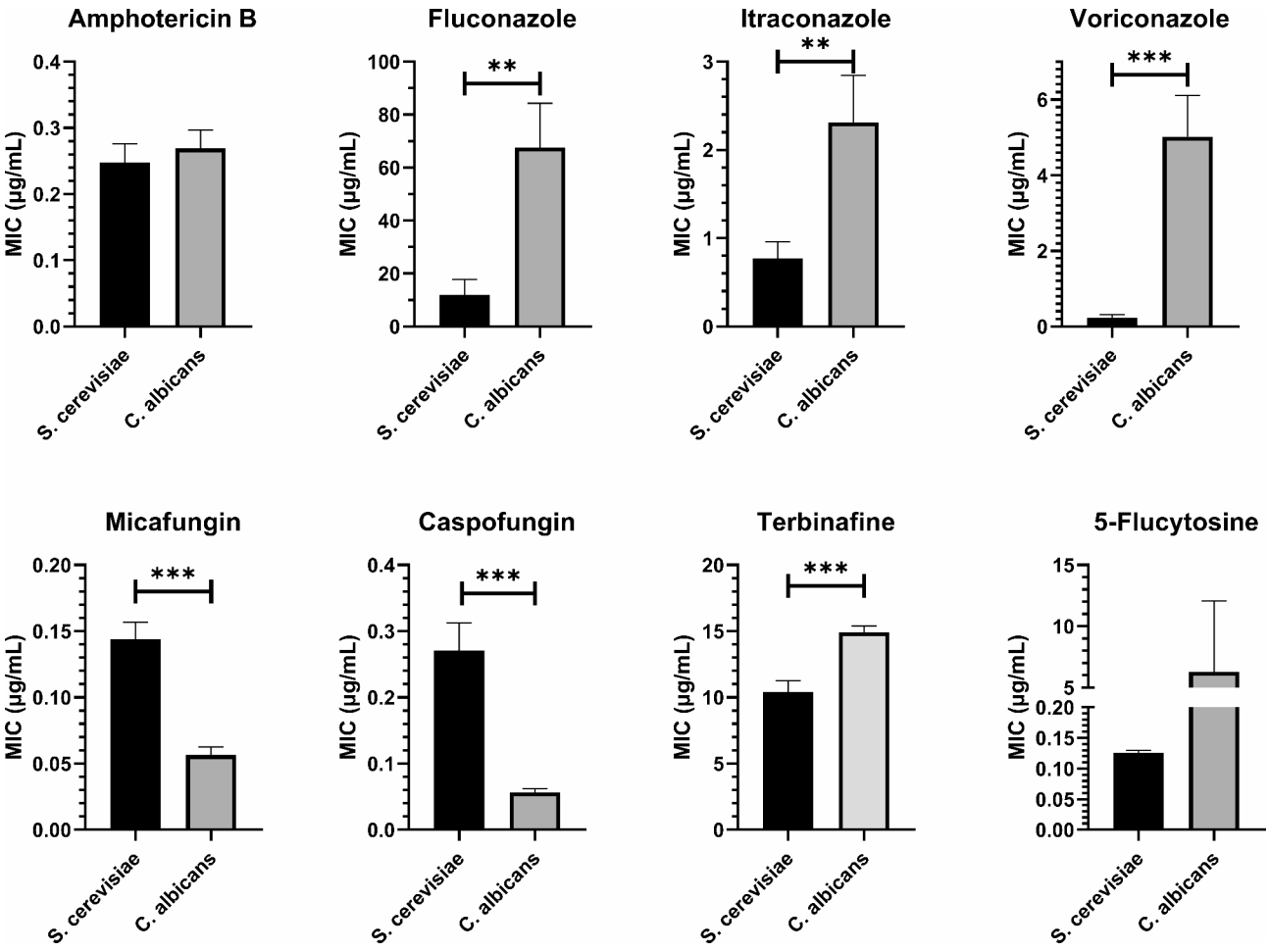

Fig. 3 Minimum inhibitory concentrations (MICs) of antifungal agents (mean ± SEM) against pathogenic fungi in vulvovaginal candidiasis (VVC). *S. cerevisiae* vs. *C. albicans* ($n=44$). **: $0.001 \le p < 0.01$; ***: $p < 0.001$

(0.60±0.15 μg/mL vs. 0.09±0.01 μg/mL, $p=0.014$), and the MIC of 5-flucytosine against isolates from the digestive system was significantly greater than that against those from the reproductive system (0.17±0.02 μg/mL vs. 0.13±0.01 μg/mL, $p=0.005$) and respiratory system (0.17±0.02 μg/mL vs. 0.13±0.01 μg/mL, $p=0.048$). The mean MICs (±Std. error of the mean) of the other antifungal agents were as follows: (1) amphotericin B: 0.74±0.44 μg/mL; (2) fluconazole: 0.74±0.44 μg/mL; (3) itraconazole: 0.60±0.15 μg/mL; (4) micafungin: 0.12±0.03 μg/mL; (5) caspofungin: 0.35±0.22 μg/mL; and (6) terbinafine: 11.61±1.26 μg/mL.

Discussion

In this study, we collected *S. cerevisiae* isolates from 75 clinical patients and calculated the MICs. To our knowledge, this is the first clinical *S. cerevisiae* study conducted in East Asia. These *S. cerevisiae* isolates were isolated from vaginal secretions, cervical secretions, ALF, sputum, feces, and intestinal mucosa, representing the reproductive system, respiratory system, and digestive system, similar to previous reports [7, 8, 12]. Given the sources of isolate transmission, *S. cerevisiae*, a yeast widely used in the food industry, is normally detected in the human digestive tract. Young children are prone to coughing while eating, with a small lung capacity, and/or iatrogenic procedures such as tracheal intubation, which may also lead to foodborne yeast aspiration into the lungs. The pathogens responsible for VVC are usually transmitted through endogenous infection, which may originate from the intestine or through colonization of the vagina. Although *S. cerevisiae* may cause invasive infections, as in previous reports [5, 8, 13], our institution has not found any evidence of invasive cases, and *S. cerevisiae* has not been detected in the bloodstream.

Few studies have reported the antifungal susceptibility of clinical strains of *S. cerevisiae*. The distributions of the MICs of amphotericin B, fluconazole, itraconazole, voriconazole and flucytosine against the clinical isolates of *S. cerevisiae* in this study (Supplementary Table 1) are consistent with those reported by Borman et al. [14]. However, compared with the study by Górzyńska et al.

Table 3 Demographic and clinical characteristics of 13 pneumonia patients with positive *Saccharomyces cerevisiae* cultures

Characteristics	No. of patients	(%)
Sex		
Male	10	76.9
Female	3	23.1
Age (years)		
< 1	9	69.2
1–4	3	23.1
5–14	1	7.7
Median age (P25-P75)	*0.50 (0.34–1.40)*	
Sample type		
Alveolar lavage fluid	11	84.6
Sputum	2	15.4
Invasive fungal disease indicators		
β-D-glucan test (G test) positive	1	7.7
Galactomannan test (GM test) positive	1	7.7
Antifungal drug use[a]		
Yes	4	30.8
No	9	69.2
Coinfection		
Bacterial infection [b]	8	61.5
Viral infection [c]	4	30.8
Mycoplasma pneumoniae infection	1	7.7
Prognosis		
Cured	4	30.8
Improved and transferred	7	53.8
Parents quit therapy	2	15.4
Exacerbation or death	0	0.0
Total	13	100.0

Note Diagnosis of pneumonia includes symptoms, signs, imaging tests and laboratory tests. (a) Antifungal drugs, including fluconazole, voriconazole and micafungin; (b) There was clear etiological evidence of bacterial infection (e.g., *Staphylococcus aureus*, *Acinetobacter baumannii*, and *Haemophilus influenzae*); (c) There was clear etiological evidence of viral infection (e.g., SARS-CoV-2, human metapneumovirus, and human bocavirus DNA/RNA amplification)

[9], which was conducted in Poland with 55 cases, the mean MICs of amphotericin B, fluconazole, micafungin, caspofungin and flucytosine differed by 2-fold, whereas the mean MICs of itraconazole and voriconazole differed by approximately 4-fold. Possible reasons for this overall difference in antifungal susceptibility include the following: (1) These studies were conducted in different regions, Eastern Europe and East Asia, and there may be large differences in the strains of *S. cerevisiae* between the two groups. (2) The race, age and sex of the subjects included in the two studies were different; this study mainly included female VVC patients, and the antifungal susceptibilities of *S. cerevisiae* isolated from different sites of the body were different.

Among the 75 clinical isolates of *S. cerevisiae* in this study, 44 were the causative pathogen of VVC. Given the heterogeneity in the susceptibility of *S. cerevisiae* isolates from different isolation sites to antifungal agents, we grouped the isolates for analysis. Another difficulty in interpreting the antifungal susceptibility of *S. cerevisiae* as a pathogen is the lack of clinical breakpoints established by scientific or industrial societies. As a medical laboratory accredited by both ISO 15,189 and the CAP, we use the CLSI guidelines to interpret the resistance of clinical isolates, including breakpoints and ECVs, but these standards lack data on *S. cerevisiae*. Therefore, to evaluate the clinical therapeutic effect of antifungal agents as realistically as possible with in vitro experiments, we used VVC patients with pathogenic *C. albicans* as a control group via stratified sampling; *C. albicans* is the primary pathogen in more than half of VVC cases.

Treatment with azoles, including topical and oral therapies, is the preferred recommended regimen for VVC according to the Sexually Transmitted Infections Treatment Guidelines (2021) [15]. In this study, the mean MICs of azole agents against *S. cerevisiae* were significantly lower than those against *C. albicans*, and the proportion of voriconazole-resistant isolates of *S. cerevisiae* was significantly lower than that of *C. albicans* with respect to the breakpoint of *C. albicans*, approximately 4.5%. In addition, after antifungal treatment, *S. cerevisiae* caused a lower rate of RVVC than did *C. albicans*, suggesting that VVC caused by *S. cerevisiae* is easier to treat and has a better prognosis.

As the resistance of RVVC pathogens to azole agents becomes increasingly common [16], treatment options other than azoles are gaining attention. In our institution, we usually treat these patients with amphotericin B vaginal effervescent tablets, to which all 88 VVC pathogen isolates in this study were sensitive according to the breakpoint of *C. albicans*. Terbinafine is an allylamine antifungal drug that inhibits squalene epoxidase in the synthesis of ergosterol in fungal cells and causes the accumulation of squalene in the cells, killing the fungus. Terbinafine is usually suitable for treating skin and nail infections caused by superficial fungi [17]. In this study, the MIC of terbinafine against *S. cerevisiae* was significantly lower than that against *C. albicans*, suggesting that it has the potential to be an adjuvant treatment for VVC caused by *S. cerevisiae*. Echinocandins (including micafungin and caspofungin) are water-soluble lipopeptides that inhibit glucan synthase and are almost exclusively available as intravenous preparations, usually for the treatment of invasive candidiasis [18]. Although there were no cases of invasive *S. cerevisiae* in this study, the MICs of echinocandins against *S. cerevisiae* were significantly greater than those against *C. albicans*, suggesting that echinocandins should be used with caution in the treatment of invasive *S. cerevisiae* infection. Flucytosine is often used in combination with other antifungal agents because resistance to this agent is common among pathogenic fungi. In this study, although the mean MIC

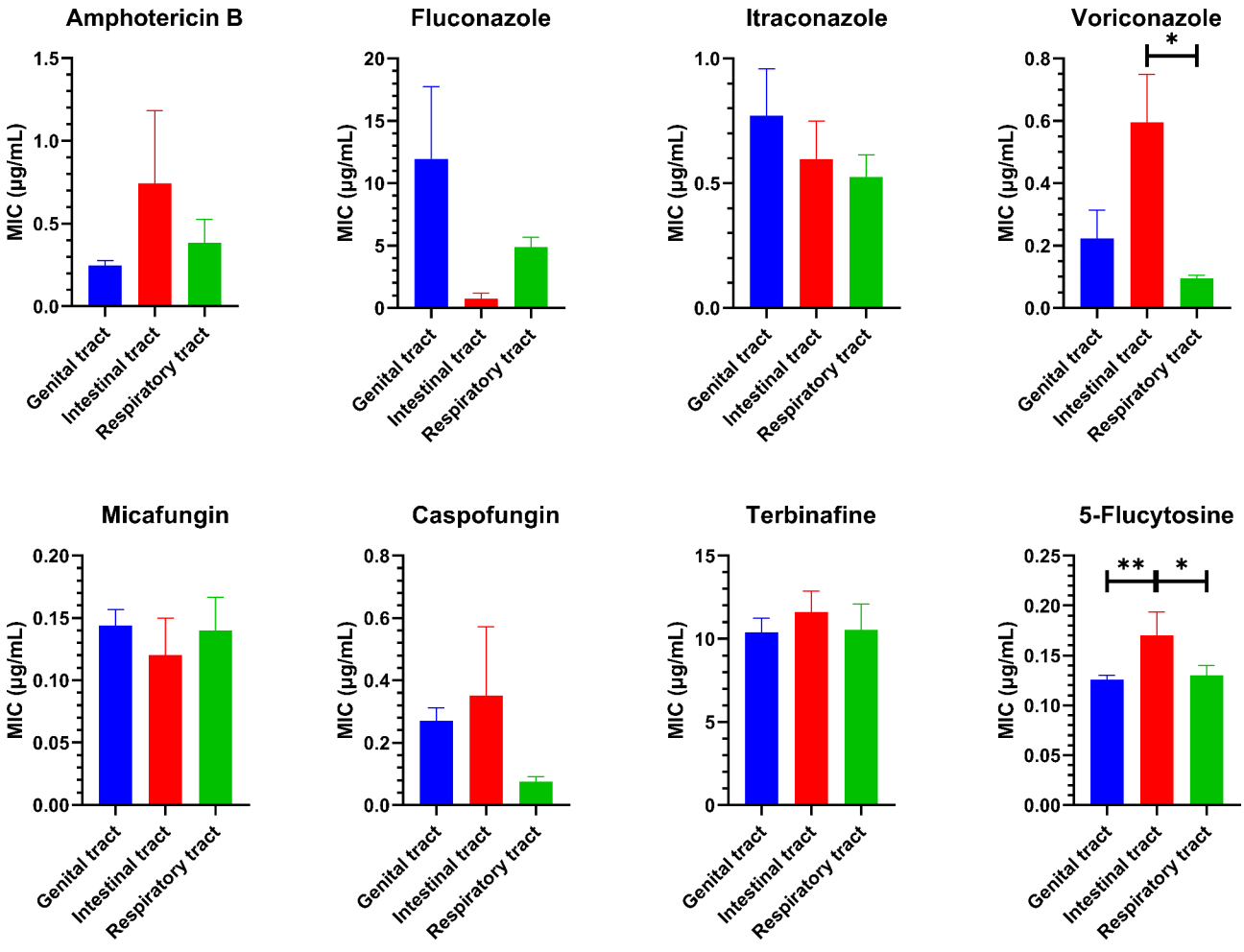

Fig. 4 Minimum inhibitory concentrations (MICs) of antifungal agents (mean ± SEM) against *S. cerevisiae* isolates from different sites. *: $0.01 \le p < 0.05$; **: $0.001 \le p < 0.01$

of flucytosine against *C. albicans* was nearly 50 times greater than that against *S. cerevisiae*, there was no significant difference between the two groups, suggesting that the efficacy of flucytosine alone is highly heterogeneous.

Compared with the study by Borman et al. [14] which also used the breakpoints of *C. albicans* to determine antifungal susceptibility, the overall resistance rates of *S. cerevisiae* to amphotericin B (0.0% vs. 0.4%), fluconazole (31.8% vs. 43.1%) and voriconazole (4.5% vs. 4.0%) in this study are similar. However, we were unable to conduct a horizontal comparison of the antifungal susceptibility of clinical *S. cerevisiae* isolates with more studies because the sample sizes, disease distributions, experimental methods, and interpretation criteria of each study varied greatly [19, 20].

In this study, we also isolated *S. cerevisiae* from children with pneumonia and diarrhea, but there was no evidence that these strains were directly related to the corresponding diseases. Studies have shown that for immunosuppressed patients, *S. cerevisiae* can cause systemic infections [6, 8, 13]. Therefore, pediatric leukemia wards in children's hospitals need to be aware of the potential pathogenicity of *S. cerevisiae*, especially in patients with pneumonia symptoms and *S. cerevisiae* isolated from the respiratory system. Moreover, *S. cerevisiae* is often made into probiotic preparations and is used orally as an adjuvant treatment for patients. Although the literature [21] indicates that *S. cerevisiae* strains are transient fungi in the intestine and do not cause colonization, some studies have shown that oral preparations may also be a channel through which *S. cerevisiae* can cause systemic infection [22, 23].

Overall, this is the first study in East Asia to report the detection and distribution of *S. cerevisiae* in medical institutions and to compare the antifungal susceptibility of *S. cerevisiae* and *C. albicans* as causative pathogens of VVC. As a newly defined rare conditional pathogenic fungus, the clinical characteristics and antifungal susceptibility of *S. cerevisiae* deserve more attention. In this study, we not only conducted an overall antifungal

Table 4 Demographic and clinical characteristics of 18 diarrhea patients with positive *Saccharomyces cerevisiae* cultures

Characteristics	No. of patients	(%)
Sex		
Male	13	72.2
Female	5	27.8
Age (years)		
< 1	9	50.0
1–4	3	16.7
5–14	6	33.3
Median age (P25-P75)	*1.17 (0.23–7.25)*	
Sample type		
Feces	15	83.3
Intestinal mucosa	3	16.7
Fecal flora dysbiosis		
Yes	9	50.0
No	9	50.0
Prognosis		
Cured	9	50.0
Improved and transferred	9	50.0
Exacerbation or death	0	0
Total	18	100.0

susceptibility analysis of clinical *S. cerevisiae* isolates but also systematically reviewed the clinical information of patients and conducted an in-depth analysis of isolates from patients with various diseases. However, this study also has several limitations. First, patient information about lifestyle and occupation was lacking (such as bread baking, frequency of body cleaning, and oral or topical use of probiotics), so we cannot determine whether the source of *S. cerevisiae* was related to lifestyle and/or occupational exposure. Second, virulence factor and molecular biological testing of the isolates was lacking, so we cannot confirm whether there are genetic differences between the pathogenic isolates and the wild-type isolates, which needs to be further explored.

Conclusion

In conclusion, medical institutions can isolate *S. cerevisiae* from the reproductive, respiratory and digestive systems of women and children, and *S. cerevisiae* can cause VVC as a pathogenic fungus. *S. cerevisiae* is less resistant than *C. albicans* to azoles, which are commonly used to treat VVC, and have a lower clinical recurrence rate. The antifungal susceptibility of *S. cerevisiae* varies according to the isolation source. Amphotericin B and micafungin may be considered when treating pathogenic azole-resistant isolates. As the detection rate of *S. cerevisiae* in medical institutions increases, attention should be given to the pathogenicity trend of the isolates, their disease prognosis and antifungal resistance should be monitored, and prevention and control of conditionally pathogenic fungi should be increased.

Supplementary Information

The online version contains supplementary material available at https://doi.org/10.1186/s12866-024-03506-y.

Supplementary Material 1

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Not applicable.

Author contributions
ZY designed the work and drafted the manuscript. YF, XT and LX completed the search and analysis of patient clinical data. JL, CM and LL completed antifungal susceptibility test and its result interpretation. XL, YC, HL, LK and YJ provided funding support and substantively revised the manuscript.

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Data availability
No datasets were generated or analysed during the current study.

Declarations

Ethical approval
The clinical experimental plan was approved by the Clinical Trial Ethics Committee of West China Second University Hospital, Sichuan University (No. 2023 – 290). Informed consent for adult participation was waived by the Clinical Trial Ethics Committee of West China Second University Hospital, Sichuan University, according to the Chinese government "Measures for the Ethical Review of Life Science and Medical Research Involving Human Beings" (Chapter III, Article 32, https://www.gov.cn/zhengce/zhengceku/2023-02/28/content_5743658.htm). For minors, informed consent was obtained from legal guardians on behalf of the minors involved in the study. The work was carried out in accordance with the Declaration of Helsinki.

Consent for publication
Not applicable.

Competing interests
The authors declare no competing interests.

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