Original Article

MEDICINE PALLIATIVE CARE

DOI: 10.47582/jompac.1325399

J Med Palliat Care. 2023;4(4):296-301

Distribution of yeasts in fungal urinary tract infections from a tertiary care hospital

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Cite this article as: Altınbaş R, Bildirici Y. Distribution of yeasts in fungal urinary tract infections from a tertiary care hospital. *J Med Palliat Care*. 2023;4(4):296-301.

Received: 11.07.2023 ◆ **Accepted**: 03.08.2023 ◆ **Published**: 30.08.2023

ABSTRACT

Aims: Urinary tract infections caused by fungi in critically ill patients steadily increasing in recent years. Fungiuria is a marker of contamination, colonization, or infection in the lower or upper urinary tract. As urinary catheters especially long-term usage was the most important risk factor for fungiuria. The identification of yeast isolates at the species level is crucial for the appropriate management of infection. We conducted this study to describe the epidemiological features of fungiuria in patients.

Methods: The yeast species were identified by using conventional methods and automated systems. Demographic data were recorded from the electronic medical records.

Results: *Candida albicans*, were predominant compared to non-*albicans Candida* species and yeast-like fungi. Among non-*albicans Candida*, the most common species were *Candida tropicalis* followed by *Candida glabrata* complex. Fungiuria was more common in females than in males. Underlying conditions were present in patients the most common risk factors were antibiotic therapy before the detection of yeasts and using a urinary catheter.

Conclusion: Epidemiological data and antimicrobial therapy play an important role in the treatment of urinary tract infections. For this reason, the identification of fungi at the species level is critical to assist the decision on antifungal therapy in complex cases. In all patients with fungal growth in urine culture, the underlying risk factors should be evaluated first. Depending on the correction of risk factors, fungiuria may resolve spontaneously. This is seen as the best approach both to reduce treatment costs and to prevent resistance to antifungals.

Keywords: Fungiuria, candiduria, urinary tract infection, urinary catheter

INTRODUCTION

Fungi and bacteria may be etiological agents in urinary tract infections (UTIs). UTIs caused by fungi in critically ill patients steadily increasing in recent years. Especially, *Candida* species important opportunistic pathogen causing UTIs.[1,2]

Candiduria (i.e., the presence of *Candida* species in urine) is a marker of contamination, colonization, or infection in the lower or upper urinary tract. There are no reference standards available for the definitive diagnosis tests to distinguish infection from colonization in urine samples.[3] Patients with candiduria can be categorized as asymptomatic or symptomatic according to diagnostic criteria.[4]

Generally, candiduria is typically rare and asymptomatic in healthy patients.[5-7] Antifungal therapy is generally not recommended in asymptomatic patients. Infectious Diseases Society of America (IDSA) strongly recommends correcting the underlying risk factors, removing or replacing the catheter in patients with urinary catheters is sufficient to prevent infection.[4,8-10]

Use of catheter, broad-spectrum antibiotic therapy, prolonged hospitalization, long duration in intensive care units (ICU), diabetes mellitus, renal disease, coronary heart disease, liver disease, prematurity, total parenteral nutrition and urinary tract malformation, immunocompromised patients are presented as more important risk factors for fungal UTI.[2,11,12] As urinary catheter especially long-term usage was believed to be the most important risk factor for candiduria.[2,8,13]

Although *Candida albicans* is the most frequently reported species in fungal urinary tract infection, non-*albicans Candida* (NAC) species (i.e. *Candida tropicalis*,

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Candida glabrata complex, *Candida parapsilosis* complex, *Candida lusitaniae*, *Candida guilliermondii*, and *Candida krusei*) are also increasing now as the causative agent of UTI. The important factor about NAC species is that they are more difficult to treat. Many strains of NAC are resistant to antifungals (i.e., many *C. glabrata* complex isolates (acquired) and all *C. krusei* isolates (adaptive) are resistant to fluconazole. Therefore, the identification of yeast isolates at the species level is crucial for the appropriate management of infection.[2,14,15]

It is important to decide in the case of candiduria whether it is colonization or an infection that requires treatment because candiduria is usually accepted as colonization or contamination by most physicians, but it may be the only sign of invasive candidiasis.[7,11,16]

At our hospital, fungal UTIs are caused by yeast such as *Candida* species and yeast-like fungi such as *Trichosporon* species. As far as we know studies on the epidemiology and prognostic value of fungiuria are not very common. Therefore, we conducted this study to describe the epidemiological features of fungiuria in patients. We aimed to evaluate the prevalence of fungi (yeast and yeast-like fungi) that cause urinary tract infections in inpatients and outpatients during a 3 years study period. In this way, we suppose that we will contribute to surveillance studies in our province, our country and perhaps globally.

METHODS

The study was carried out with the permission of Eskişehir Osmangazi University Non-interventional Clinical Researches Ethics Committee (Date: 26.10.2021, Decision No: 10). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

In this study, we evaluated the results of all urine samples taken from the patients who applied to our laboratory with a preliminary diagnosis of urinary tract infection for three years (2019-2021).

The urine samples were collected and stored in a disposable sterile bottle with a screw lid. Before sample collection, patients were instructed on how to collect mid-stream urine.[17] All urine samples received from one patient were considered as a single sample. Although urine culture is regarded as the gold standard for the detection and identification of fungi, microscopic examination of urine is a useful tool for diagnosing fungal UTI.[18] Microscopy of yeast in the urine such as budding cells, ovoid- shaped yeast cells, hyphal element (pseudohyphae) and arthroconidia is the first clue to the presence of a fungal infection.[19]

Urine specimens were inoculated on blood agar (RTA, Turkiye), Eosin Methylen-blue (EMB) agar (RTA, Turkiye) and Sabouraud's dextrose agar (SDA) (RTA, Turkiye) with a calibrated loop technique delivering 0.001 ml of urine as per standard protocol for urine culture. The culture plates were incubated aerobically at 35°C for 24-48 h. The yeast species were identified by using of tests including Gram's staining, germ tube test, chlamydospore formation on corn meal agar (RTA, Turkiye), color appearance on CaandidaCHROM agar (RTA, Turkiye) and VITEK 2 Compact (bioMérieux, Marcy l'Etoile, France) identification system which is YST card was used for yeast and yeast-like fungi (YLF).

Demographic data including age, gender, comorbidities, cause of admission (medical or surgical) and the underlying malignancy were recorded from the electronic medical records. Likewise, previous antibiotic use and urinary catheter insertion were recorded from electronic medical records.

RESULTS

During the study period, we received 61843 urine samples from different polyclinics, wards, and ICUs of our tertiary city hospital. 10187 (16.47%) were found positive; among them, 9009 (88.44%) and 1178 (11.56%) had bacteriuria and fungiuria, respectively. Regarding the urine culture results of our patients, growth was not detected in 46073 (74.5%) of the patients and contamination was detected in 5583 (9.03%).

In total, 1183 strains of yeasts isolates were recovered from the 1178 patients with fungiuria. The distribution of species is shown in **Table 1**. *C. albicans* (n=557 47.1%), were predominant compared to NAC species and YLF. The incidence rate of yeasts was determined as *C. tropicalis* (n=230 19.4%), *C. glabrata* complex (n=133 11.2%), *Trichosporon asahii* (n=70 5.9%), *C. parapsilosis* complex (n=41 3.5%), *Candida kefyr* (n=41 3.5%). *C. lusitaniae* (n=22 1.9%), *C. krusei* (n=18 1.5%), *Candida famata* (n=16 1.4%), *Trichosporon mucoides* (n=12 1.0%) and other *Candida* species (n=43 3.6%).

In our study, NAC species were isolated in 46.0% and YLF were isolated in 6.9%. Among NAC, the most common species were *C. tropicalis* followed by *C. glabrata* complex. Among YLF, the most common species were *T. asahii* (5.9%) followed by *T. mucoides* (1.0%).

Mixed infections with two microorganisms were found in thirty-five cultures (3.0%), thirty (2.6%) together with bacteria and five (0.4%) with two different yeasts (**Table 2**).

Table 1. Percentage of distribution of urinary isolates of fungi species

Species	No	%
Candida albicans	557	47.1
Candida tropicalis	230	19.4
Candida glabrata complex	133	11.2
Candida kefyr	41	3.5
Candida parapsilosis complex	41	3.5
Candida lusitaniae	22	1.9
Candida krusei	18	1.5
Candida famata	16	1.4
Other *Candida* Species*	43	3.6
**Candida guilliermondii*	9	20.9
**Candida ciferrii*	9	20.9
**Candida dubliniensis*	6	14.0
**Candida melibiosica*	5	11.6
**Candida lambica*	4	9.3
**Candida sphaerica*	4	9.3
**Candida pelliculosa*	2	4.7
**Candida rugosa*	2	4.7
**Candida norvegensis*	1	2.3
**Candida utilis*	1	2.3
Trichosporon asahii	70	5.9
Trichosporon mucoides	12	1.0
Total	1183	100.0

Table 2. Mixed infections of yeasts and bacteria found in urine with fungiuria

Mixed infections	number	%
With bacteria	30	85.7
Candida albicans+*Enterococcus faecium*	17	48.6
Candida albicans+*Enterococcus faecalis*	1	2.9
Candida albicans+*Staphylococcus hominis*	1	2.9
Candida albicans+*Escherichia coli*	1	2.9
Candida albicans+*Klebsiella pneumoniae*	1	2.9
Candida albicans+*Pseudomonas aeruginosa*	1	2.9
Candida tropicalis+ *Enterococcus faecium*	1	2.9
Candida tropicalis+*Enterococcus faecalis*	2	5.7
Candida tropicalis+*Morganella morganii*	1	2.9
Candida tropicalis+*Staphylococcus hominis*	1	2.9
Candida glabrata complex+*Enterococcus faecalis*	1	2.9
Candida glabrata complex+*Acinetobacter baumannii*	1	2.9
Candida glabrata complex+*Klebsiella pneumoniae*	1	2.9
With two yeasts	5	14.3
Candida albicans+*Trichosporon asahii*	3	8.6
Candida albicans+*Candida glabrata* complex	1	2.9
Candida tropicalis+*Candida kefyr*	1	2.9
Total	35	100.0

Out of these 1178 fungiuria cases, 793 (67.3%) patients were from ICU, 292 (24.8%) were from wards and 93 (7.9%) were from polyclinics. *C. albicans* was the most frequently isolated species from the samples of patients in the ICUs (365/793 46.0%) and in the wards (132/292 45.2%).

In this study, fungiuria was more common in females 660 (56%), than in males 518 (44%). Out of 1178 isolates, 93 (7.9%) were from outpatients and 1085 (92.1%) were isolated from inpatients.

The mean age of presentation of the patients was 68.34±17.69 years (range 0-96 years). Age group analysis showed that 31 (2.6%) patients were under 16 years old, 32 (2.7%) were between 16 and 30 years, 125 (10.6%) were between 31 and 55 years, 496 (42.1%) were between 56 and 75 years, and 494 (41.9%) were older than 76 years (Table 3). The highest isolation rates of yeast were found in an age group above 55 years.

Table 3. Age and sex distribution pattern of fungi species from urine samples (n=1178)

Age	Male	Female	No %
<15	13	13	26 (2.2)
16-30	5	32	37 (3.1)
31-55	42	83	125 (10.6)
56-75	240	256	496 (42.1)
>76	218	276	494 (41.9)
Total	518 (44%)	660 (56%)	1178 (100%)

Less than half of the patients (445/1178 37.8%) had comorbidities, the most common of which were diabetes mellitus 7.81% (n=92), lung diseases 7.30% (n=86), hypertension 4.75% (n=56), malignant neoplasm 4,41% (n=52), vascular and heart diseases 4.16% (n=49), kidney diseases 3.99% (n=47), cerebrovascular diseases 2.29% (n=25) and immunodeficiency 1.1% (n=13). Ten patients had kidney stones, nine patients had urinary tract anomalies, three patients had cirrhosis and three patients had vaginitis. Our study showed that candiduria is common in patients with diabetes mellitus.

Underlying conditions were present in patients the most common risk factors were antibiotic therapy (998/1178 84.7%) before the detection of yeasts and using a urinary catheter (860/1178 73%). Of the patients using urinary catheters, 86.6% (n=745) were hospitalized in the intensive care unit and 13.4% (n=115) were hospitalized in wards.

DISCUSSION

Epidemiological data and empirical antimicrobial therapy play an important role in the treatment of urinary tract infections, which cause a significant burden worldwide. For this reason, the identification of fungi at the species level is critical to assist the decision on antifungal therapy in complex cases.[20] Our study characterizes a single-center experience on fungiuria over three years. The presence of a wide variety of fungi (yeasts and YLF) in the urine is known as fungiuria. The presence of *C. albicans* and NAC species in the urine is known as candiduria.[7]

Even though NAC species now account for a significant proportion of clinical isolates collected worldwide in hospitals. *C. albicans* is the most common cause of nosocomial fungal urinary tract infections. Regarding the high resistance to some antifungal agents of NAC species such as *C. glabrata* complex and *C. krusei*, the detection of NAC species in patients' urine samples should be important for the treatment.[21]

Contamination is common in patients who do not follow clean urine specimen collection guidelines. A new urine sample is recommended to exclude contamination. If the new specimen yields no yeasts, there is no need to continue studies for diagnosis.[22]

Fungus is considered to be the second leading pathogen causing UTI in ICUs after *Escherichia coli*.[23] Fungal UTIs encompass a broad variety of fungi. The overwhelming majority of UTIs are caused by *Candida* spp. but yeast-like species are also prevalent such as *Trichosporon* spp.[19]

The *C. albicans* was the most important yeast associated with human candiduria in the last decades.[21] The reported incidence of candiduria varies (10-68.42%) in different geographical locations. In the literature, *C. albicans* growth was reported to vary widely.[8,10,11,13,14,20,24-27]

In the present study, the frequency of *C. albicans* (47.1%) was higher than NAC species (46.0%) and yeast-like species (6.9%). Among NAC species, *C. tropiclis* (19.4%) followed by *C. glabrata* complex (11.2%) had the highest frequency compatible with the literature.[2,28] Contrary to a study by Paul et al.[29] and Kobayashi et al.[10] according to our study *C. albicans* is the most common species of fungiuria. Similar to some previous studies, we found that *C. tropicalis* was the most common of the non-*albicans* species isolated.[2,21,25] It was reported as *C. tropicalis* (22.2%) was the most common of the non-*albicans* species isolated and non-*albicans* yeasts were found at 42.2% in another similar study.[10] Nevertheless, our data was found to differ from the data presented by Singla et al.[30] and Pramodhini et al.[25] They reported that *C. tropicalis* has been frequently (first leading agent) identified in 57.3% and 65.7% of patients with candiduria respectively. Although lower than the results of the two researchers, Paul et al.[29] found that *C. tropicalis* was the most frequently isolated microorganism from candiduric patients with an incidence rate of 30.5%, followed by *C. albicans*.[3]

Although in our study and some previous studies *C. glabrata* complex was found to be the third leading agent of candiduria, generally the incidence of *C. glabrata* complex isolated from urine is increasing throughout the world.[6,13,15,17,31,32] There is a consensus in some studies that the high prevalence of NAC species, especially *C. glabrata* complex, increases in diabetic patients.[17,31] *C. parapsilosis* complex uncommon in urine and frequently significantly lower than the *C. trpicalis* and *C. glabrata* complex in our study. This result is also compatible with the literature.[6,7,15] Our opinion associated this with the low number of neonates and pediatric patients in our study.

In the present study, *T. asahii* is the most prevalent species among the yeast-like strains isolated from urine samples. However, in contrast with that Gharaghani et al.[3] reports showed *Trichosporon* species was more commonly isolated from the urine samples but our study similar to Francisco et al.[19]

In our study, we detected *Candida* species simultaneously with another urinary tract pathogens (yeast-bacteria) or two different yeast species (yeast-yeast) in the same urine samples.

In total, five patients had mixed infections caused by two different yeast and 30 patients had mixed infections caused by bacteria (**Table 2**). Mixed infections of yeasts and bacteria were found in the urine with fungiuria; the *C. albicans* 4.7% co-isolated with other species, *C. glabrata* complex 2.7% co-isolated with other species and *C. trpicalis* 2.6% co-isolated with other species. Gharaghani et al.[21] found similar results in their study.

In some studies, the frequency of yeast-yeast or yeast-bacteria co-isolation in candiduria cases has been reported as (3.5-10%).[1,3,11,12,26,33] The data obtained from our study revealed that 3.0% of candiduria patients can harbor two or more species at the same time. According to the results of many studies, our finding was found to be low.[11]

The incidence of candiduria in our hospital was most common in patients admitted to ICUs (67.3%). Similarly, according to different studies, the most important predisposing factors in creating candiduria were hospitalization in ICUs.[12,14] We think that this may be related to the prolonged stay in the ICU, the increase in the number of interventional procedures and the long-term use of empirical antibiotics.

In our study the female:male ratio (1.3:1) was observed similar to the female dominance in previous studies. Since *Candida* species are an important part of the normal microbial flora in the vagina, many researchers associate the high frequency of candiduria in women with vulvovestibular colonization and vaginal candidiasis.[34] Three of the women (3/660 0.45%) had vulvovaginal candidiasis in our study. Many researchers associate, women are more often affected due to their shorter urethra.[3,6,10,13,17,25,26,31] Several studies have shown male predominance in the incidence of candiduria which are in disagreement with our study.[3,8,14,35]

The incidence of candiduria may vary according to age groups. According to previous studies, the incidence of candiduria was significantly increased when compared with the overall incidence in patients over 60 years of age and under 1 year of age.[25]

In our study, the majority of patients identified with fungiuria were in the age group of 56 to 75 years (42.1%), followed by the 76 to 96 age group (41.9%). Our study has shown a high prevalence rate of UTIs in elderly people, which is concordant with other studies. Similar to many studies, in our study we noted the most common risk factors for fungiuria: age older than 55 years, female sex, diabetes mellitus, use prior antibiotics, catheter and ICU stay.[15]

We found several risk factors, diabetes in 7.81%, lung diseases in 7.30%, hypertension in 4.75%, malignant neoplasm in 4.41%, vascular and heart diseases in 4.16%, kidney diseases in 3.99%, cerebrovascular diseases in 2.29% and immunodeficiency in 1.1%. Diabetes mellitus was the most common underlying disease seen in the patients. It was found to be similar to previous studies.[15,33]

Once the presence of *Candida* in the urine is confirmed careful clinical evaluation should be performed to detect symptoms, diabetes mellitus, genitourinary structural abnormalities, decreased renal function and metabolic syndromes.[18] Patients with diabetes are at increased risk for candiduria, glycosuria and acidic pH can be due to an increased susceptibility to *Candida* colonization rates.[13,31] In conditions of immune deficiency, the commensal fungal microorganisms may convert into opportunistic pathogenic microorganisms, creating fungal UTIs.[34]

In many studies, it is stated that candidemia develops with candiduria in the presence of obstruction or urinary tract instrumentation, however in a low percentage.[17] Candidemia was not observed in any of the patients included in our study.

Catheters serve as a portal of entry for yeast into the urinary system.[3,33] Catheter-associated urinary tract infection represents one of the most common healthcare-associated infections in patients.[36] The presence of a catheter plays a crucial role in the pathogenesis of candiduria; biofilm formation and migration of organisms along the surface of the catheter to organism persistence.[13,17,18,33] *C. albicans* and *C. tropicalis* are the species with the highest adhesion and biofilm formation ability on the urinary catheter. Biofilm is a virulence factor and biofilm affects the development of antifungal resistance.[37] It should be noted that these are the most frequently isolated species in our study.

Study Limitations

The first limitation of this study is that it was performed with a retrospective design. Second, the sample size was small and finally, antifungal susceptibility testing of the isolated yeast species was not performed, thus providing no information on fluconazole resistance trends in isolated fungal strains.

CONCLUSION

In all patients with fungal growth in urine culture, the underlying risk factors should be evaluated first. Depending on the correction of risk factors, fungiuria may resolve spontaneously. This is seen as the best approach both to reduce treatment costs and prevent resistance to antifungals.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Eskişehir Osmangazi University Non-interventional Clinical Researches Ethics Committee (Date: 26.10.2021, Decision No: 10).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

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