ISSN: 2833-4353

Research Article

Archives of Epidemiology & Public Health Research

Species of Yeasts Identified from Different Clinical Samples Addis Ababa, Ethiopia

Elias Seyoum[1*]

[1]*Ethiopian Public Health Institute, Addis Ababa, Ethiopia*

***Corresponding Author**
Elias Seyoum, Ethiopian Public Health Institute, Addis Ababa, Ethiopia, eliasseyoum2@gmail.com

Submitted: 21 Oct 2022; **Accepted:** 31 Oct 2022; **Published:** 16 Nov 2022

Citation: *Elias Seyoum (2022). Species of Yeasts Identified from Different Clinical Samples Addis Ababa, Ethiopia. Archives of Epidemiology & Public Health Research. 1(2), 73-79.*

Abstract

Background

Currently, fungal diseases in humans are among the most problematic illnesses. This is the result of an increase in opportunistic fungal infections caused by a rise in the number of immune-compromised patients. Particularly, yeasts are the most common cause of fungal infections leading to a range of superficial types to life-threatening invasive infections. The aim of this study is to determine the species distribution of yeasts particularly of Candida species recovered from different clinical samples.

Method

A cross sectional study was conducted from January 2018 to September 2018 in Addis Ababa, Ethiopia. Specimen from Oropharyngeal, nail, eye's cornea, and vaginal were collected from patients with signs and symptoms of infections. Identification was performed by using a YST card through VITEK 2 compact system. All data were coded, double-entered and analyzed using SPSS version 20.

Result

174 yeasts were recovered of which 51.1% were Candida albicans, 43.1% were nonalbicans Candida species and 5.7% were yeasts other than Candida species. Eighteen (18) different types of yeast species were identified from different clinical samples. All recognized yeast is considered as a causative agent of fungal infection in previously reported data worldwide.

Conclusion

C. albicans remaining the predominant species, but the emergence of non-albicans Candida and other than candida species have increased. Thus, more studies on Candida prevalence are needed throughout country.

Keywords: *Candida Albicans, Non- Albican Candida Species, Candidiasis.*

Abbreviation:
SPP: Species
C: Candida
DRERC: Department of research and ethical review committee;
TSY: Trypticase soya broth;
ATCC: American Type Culture Collection;
ISO: International Organization for Standardization.

Background

Currently, fungal diseases in humans are among the most problematic illnesses to manage. Some fungi cause disease in immune-competent persons (fungal true pathogens), but most fungal infections occur in immune-compromised individuals (opportunistic fungal pathogens). Since risk factors for opportunistic fungal infections continue to increase in frequency, it is likely that the incidence of opportunistic fungal infections will continue to increase in the future. An increase in opportunistic fungal infections is the result of an increase in the number of immune-compromised patients, thereby threaten the achievement of the newest medical advances in cancer care, solid organ and hematopoietic stem cell transplantation, neonatal medicine, autoimmune disease therapies, trauma and intensive care, and sophisticated surgery [2]. Fungi can cause infections ranging from easily treatable superficial type to life-threatening invasive infections and it has the capability to infect humans of all age groups [3]. Particularly, yeasts are the most common cause of fungal infections, leading to a range of life-threatening invasive diseases such as blood stream Candidiasis, pneumonia, and *Cryptococcal* meningitis to non-life-threatening mucocutaneous Candidiasis such a, vulvovaginal Candidiasis, and oropharyn-

geal Candidiasis [4]. They are also important cause of superficial mycosis such as onychomycosis [5]. Among fungal infections, blood stream Candida infection (candidemia) and Cryptococcosis are commonly associated with high morbidity and mortality rate. For example, Candida species are among the top ten pathogens causing bloodstream infections resulting in significant increase in the length of patients' hospitalization and in healthcare costs [4].

Currently, there are more than 150 known species of Candida [3], and more than 17 different Candida species are known as etiological agents of human infection [13]. Among Candida, *Candida albicans* is the most common infectious agent. However, there has been an important shift to non-albicans Candida species. The aim of this study was to determine distribution profile of yeasts particularly of Candida species recovered from different clinical samples collected from patients referred for culture testing by using the VITEK 2 compact system.

Methods and Materials

Study Area, Study Design and Period

The study was conducted at Arsho Medical laboratory, Addis Ababa, Ethiopia. A cross sectional study was conducted from January 01, 2018 to September 30, 2018.

Sample Collection

All samples were collected according to standardized operating procedures. Clinical samples including oropharengial swabs, nail scrapings, corneal scrapings, and vaginal swabs were collected from patients with signs and symptoms of infections and referred to the study site for culture and susceptibility testing. A portion of each clinical specimen was inoculated onto bacteriological culture media for routine activity and incubated at the appropriate temperature and period according to standard protocols related to each sample. The other portion of each clinical samples were inoculated on to Sabouraud dextrose agar (Oxoid, Basingstoke, UK) to which 50µg/1ml gentamicin is incorporated. Inoculated sample were kept at least 72 hours a temperature of 370 C. Yeast isolates were transferred to tryptic soya broth with 20% glycerol and transport to the Ethiopian Public Health Institute National reference laboratory of Clinical Bacteriology and Mycology case team, which is an ISO 15189 accredited laboratory, and stored at minus 80°C deep freezer until used.

Identification

Identification testing was performed by YST card through VITEK 2 system. A study in 2007 demonstrated of 750 clinical yeast isolates to check the performance of VITEK 2 YST identification card was evaluated by compared to the RapID Yeast Plus system using 16S rRNA sequence analysis was used as the reference method ,98.2% of isolates were correctly identified to the species level by the VITEK 2 system [14], So this study confirmed VITEK 2 system is rapid and accurate methods for identification of yeast species [2].

The viability of yeast original cultures were checked by plating yeast colonies on Sabouraud dextrose agar (Oxoid, Basingstoke,UK). Yeast identification testing were determined by the automated VITEK 2 compact system (bioMérieux, France) using YST-21343. The inoculums suspensions for the VITEK 2 were prepared in sterile saline at a turbidity equal to a 2.0 McFarland standard, as measured using a Densichek instrument (bioMérieux). The YST-21343 were automatically filled with the prepared culture suspension, sealed, and incubated by the VITEK 2 instrument. The cards were incubated at 35.5 °C for 18 h, and data were collected at 15-min intervals during the entire incubation period and final identification testing results were obtained in approximately 18 h or less. The final profile results were compared with the database, and the identification of the unknown organism was obtained. A final identification of excellent, very good, good, acceptable, or low- discrimination was considered correct.

Quality Assurance

Expiry date of media, VITEK card of YST, sterility and performance of media and VITEK card checked and a standard protocol was followed. Specimens were collected following SOPs and processed in Arsho microbiology laboratory. Isolates were transported to the laboratory of Ethiopian public health institution national reference laboratory of clinical bacteriology and mycology as soon as possible after identify isolates for further identification test. Pre-test was done before regular data collection was started. For performance, check VITEK 2 Compact system tested by using Standard strains of *Candida albicans* ATCC 10231 before any isolate tested. Reference strains for quality control were passed. Quality of the data were maintained by coding the isolates with unique number and finally all clinical isolates were preserved in deep freeze using 20% glycerol with trypticase soya broth (TSY) in case needed or for future further investigation. Data were checked for completeness before analysis.

Data Processing and Analysis Results were compiled and entered in to SPSS version 20 Software and were analyzed to determine Frequency, percentage and compare distribution of each etiologic agent by anatomic site of infection was calculated involved.

Ethical Consideration

All ethical considerations and obligations duly addressed, and the study conducted after the approval of the department of research and ethical review committee (DRERC) of the department of Medical Laboratory Sciences. Written informed consent obtained from the participants before data collection. Each respondent provided the right to refuse to take part in the study and to withdraw at any time during the study period. All the information obtained from the study subjects coded to maintain confidentially. When the participants found to be positive for fungal pathogen, they informed to the hospital clinician and received proper treatment. An assent form completed and signed by a family member and/or adult guardian for participants under the age of less than 18 years.

Results

A total of 645 samples from different body sites, which consisted of 200 vaginal, 151 corneal, 194 nail and 100 Oropharengial were

collected and evaluated for yeast species distributions. As shown in Table 1, 174 yeasts recovered of which 51.1% (89/174) were *Candida albicans*, 43.1% (75/174) were non-albicans Candida species and 5.7% (10/174) were yeasts other than Candida species. From 174 isolates were identified from different clinically suspect a fungal infection, the frequency of isolate from vaginal specimen highest 50% (87/174) followed by Oropharngial 28.7% (50/174), Nail 14.3% (25/174) and Eye 6.9% (12/174).

Yeast species with major Candida species identified in different quantity of distribution. The most commonly isolated species were: *Candida albicans* 51.1% (89/174) followed by *Candida krusei* 7.5 % (13/174), *Candida famata* 6.9% (12/174),*Cryptococcus laurentii* 4% (7/17),*Candida rugosa* 2.2%(,4/174), *Candida lusitaniae* 5.2% (9/174),*each of Candida parapsilosis and Candida lipolytica* 3.4%(6/174), each of *Candida dubliniensis* ,*Candida kefyr, Candida guilliermondii* 2.9%(5/174) , *each of Cryptococcus neoformans* and *Candida ciferrii* 1.1% (2/174) *and Candida pelliculosa* 2.3% (4/174) and other yeast species 2.8% (5/174).

Candida albicans were the main isolate in all specimen type whereas in nail the highest isolates were Candida famata followed *Candida albican and Cryptococcus laurentii* with equal frequency
C. krusei 17.1% (13/75), *C. famata* 16 % (12/75), and *C. lusitaniae* 12% (9/75) were the commonest isolates among non-albican candidia species. *Candida krusei* the main isolated in vaginal and C. lusitaniae and *Candida famata* from all specimen recovered. Whereas, yeast fungal infections of the eye showing highest variability of non-albican candida species with equal frequency [Table1].

Table 1: Species distribution of yeasts isolated from different clinical samples from January 2018 to September 2018 in Addis Ababa, Ethiopia.

	Clinical samples				
Species	**Vaginal Discharge**	**Oropharyngeal**	**Nail**	**Eye Discharge**	**Total**
Candida albicans	52	28	4	5	89
Sub-total					
Non-albicans candida					
C. krusie	11	0	0	2	13
C .famata	2	4	5	1	12
C. guilliermondii	0	3	1	1	5
C. lipolytica	2	4	0	0	6
C .pelliculosa	1	0	3	0	4
C. intermedia	0	0	1	0	1
C. utilis	0	1	0	0	1
C .rugosa	0	2	1	1	4
C. glabrata	2	0	0	0	2
C. lusitaniae	3	2	3	1	9
C. kefyr	1	2	1	1	5
C. dubliniensis	5	0	0	0	5
C. parapsilosi	2	4	0	0	6
C. ciferrii	2	0	0	0	2
Sub total					
Other yeast					
Cryptoccocus laurenti	3	0	4	0	7
C. neoformans	1	0	1	0	2
Trichosporn mucoides	0	0	1	0	1
Sub-total					
Grand Total	87	50	25	12	174

A non Candida yeast isolates from different clinical samples profile were *Cryptococcus neofor mans, Trichosporn mucoid, Cryptococcus laurenti.* Among yeasts other than Candida species *Cryptococcus laurenti* represented 70 % (7/10) this group of yeasts.

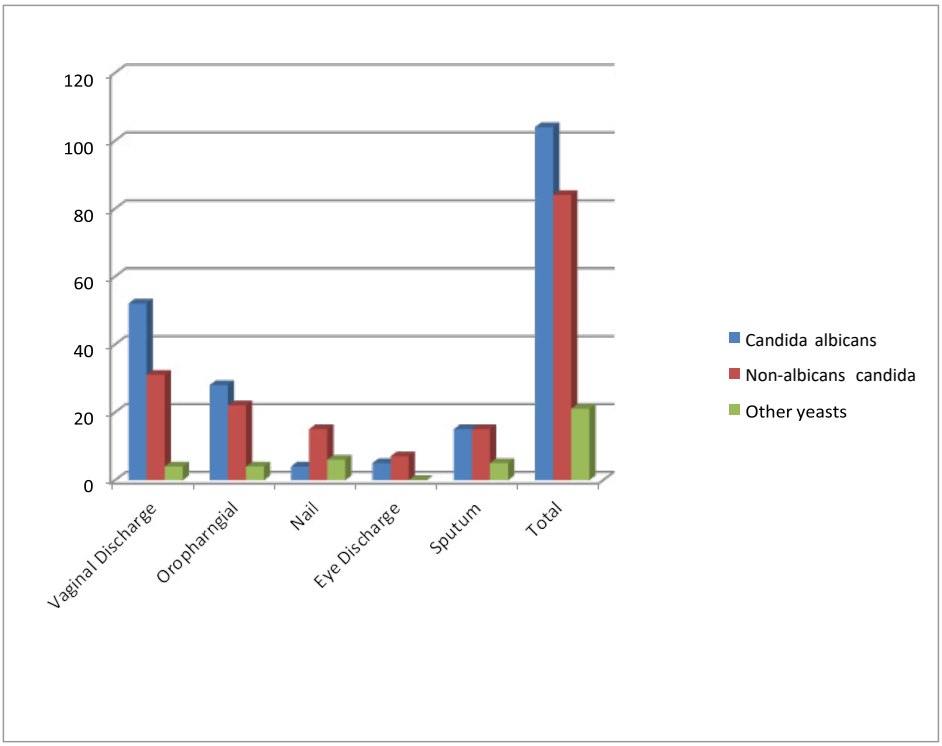

Figure 1: Frequency of *Candida albican, non-albicans Candida* and other yeasts species from different clinical samples in Addis Ababa, Ethiopia from January, 2018 to September.

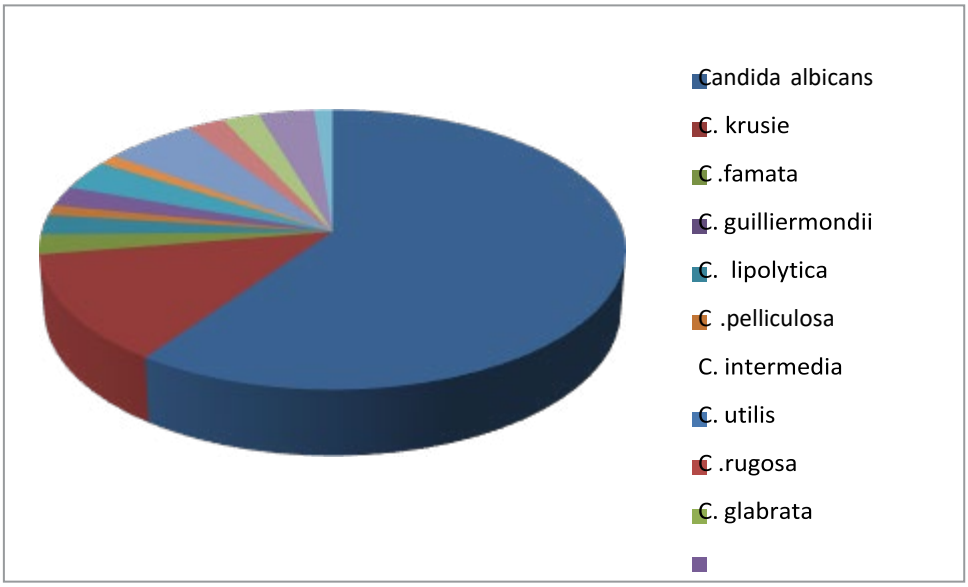

Figure 2: Species distribution of yeasts isolated from different clinical samples from January 2018 to September 2018 in Addis Ababa, Ethiopia.

Discussion

The incidence of opportunistic fungal infections such as Candida infections has considerably increased recently. Given the fact that many clinical microbiology laboratories in Ethiopia do not perform identification of yeasts beyond direct microscopy, accurate identification of Candida down to the species level is crucial. This is because different species have different antifungal susceptibility profiles and the incidence of non-albicans candida is increasing. In the present study, the identification of yeasts down to the species level profile was determined in an automated manner by the VITEK 2 compact system that is reducible and precise when compared with other reference methods [8-10]. Although the relative prevalence of the yeast species depends on the geographical location, patient population, and clinical settings [65], with a rise in immunosuppressive patients, an increased number of fungal infections is reported worldwide.

Simultaneously, the profile of human yeast pathogens has also been increasing [17]. Whereas, In Ethiopia little is known regarding the distribution and the in vitro antifungal susceptibility profile of yeasts isolated from patients. The different types of yeast species identified from clinical suspect vulvovaginal, oro-

pharyngeal, sputum, nail and eye fungal infection patient with major Candida species of yeast. These are *Candida albicans, Candida glabrata, Candida parapsilosis, Candida krusei, Candida guilliermondii, Candida lusitaniae, Candida dubliniensis, Candida pelliculosa, Candida kefyr, Candida intermedia, Candida lipolytica, Candida famata, Candida rugosa, and Candida pelliculosa, Creptococus laurentii, Creptococus neoformans, Trichosporn mucoides, Candida ciferrii.* This result demonstrated a similar finding with previously published data, which was reported considered as a causative agent of fungal infection, worldwide [11, 6, 7].

There is no recognized information regarding the profile of yeast species from Ethiopian patient's similar results with this data. In the present study, 43.1% were non-albicans candida species, one report in Addiss ababa, Ethiopia from a vaginal specimen demonstrated isolate of Candida species data showed similar to this result [9] [12]. The ratio between *C. Albicans* to non-albicans Candida species was being 1.2 to 1. Among non-Albicans Candida spp. *C. krusei (13), C. famata (12),* and *C. lusitaniae (9)* were the predominant species. *C. albicans* and *C. krusei* as the "1st and 2nd" predominate species were similar with a study conducted in Ethiopia [9] [12], but *C. famata* and *C. lusitaniae* two of the dominant species in the current study were not recovered at all by previous Ethiopian study [12], as well as in the world consider as rare finding the of human pathogen [6].

Occurrence rate of *Candida rugosa* 2.2 % (4/174) isolated from oropharengial, isolate followed by coneal discharge and nail. These data result was higher while compare to Ghana isolates reported 0.7% of total Candida isolates (4/600), which is similar to the reported incidence of 0.6% worldwide. The variation arise may be due to regional and patient population of in this study [19] [13]. Clinically, species of the *C. rugosa* complex have been isolated from a range of sources including blood, urine, sputum, and swabs from different anatomical sites [20]. Members of the *Candida rugosa* species complex have been described as emerging fungal pathogens and are responsible for a growing number of Candida infections [21]. Studies are needed to better clarify the frequency of *Candida* rugosa infections in Ethiopian patients.

50 yeasts were recovered from 100 oropharngial specimen with predominant *Candida albican* 56 % (28/ 50) isolated. It has great variation previously reported data in Ethiopia 81% [18] and Ghana 76.19% from HIV patients [15]. However, one study in China from patients in intensive care units (ICU) result comparable *Candida albican* (40.1%) to this finding [11].This deviation possibly arises from the study population difference and low number of sample size. In oropharengial, the incidence rate of non-albican candida yeasts were around 44% (22/50) and other than non-Candida yeast was not found which is similar with previously study [18, 10]. 6.9 % (12/174) from total of isolate recovered from eye discharge with main isolate of *Candida albican* and high variability of non-albican Candida species with equal frequency of each 0.5% (1/174) that were *C. lusitaniae, C. kefyr,C. rugosa,C. famata,C. krusi* and *C. guilliermondii.* Fungal keratitis is one of the most challenging types of infectious keratitis, which has been gradually increasing during the past few decades. It now accounts for approximately 50% of infectious corneal diseases [22].

In the current study, the occurrence rate of non-candida yeast isolates were 5.7 % with highest frequency of *Cryptococcus laurentii* 4 % (7/174) followed by *Creptococcus neoformans* 1.1% (2/174) and *Trichosporn mucoides* 0.5% (1/209). *Cryptococcus laurenti and Trichosporn mucoides* are a rare human pathogen, but Creptococcus neoformans as typical fungal pathogen to immunocompromised [13]. When comparing these data of *Cryptococcus laurentii* 4 % to previously one study showed 0.6 % (1/155) recovered rate in Ethiopia from oropharyngeal [23].

All epidemiological papers of fungal diseases in Ethiopia were reviewed. Where there was no Ethiopia data align with this finding. Perhaps, by reason of this diseases are often understudied. *Cryptococcus laurentii* generally cause superficial to deep-seated infections in immunosuppressed patients. Other than *Cryptococcus neoformans* species have classically been considered to be non-pathogenic. However, *Cryptococcus albidus, Cryptococcus laurentii, Cryptococcus luteolus Cryptococcus uniguttulatus, Cryptococcus curvatus,* have emerged as opportunistic pathogens over the last few years [24]. They have been described as opportunistic pathogens in HIV positive individuals, as well as in patients with other predisposing factors [25]. *Creptococcus laurentii* has been implicated in 18 cases of opportunistic infection, predominantly of the skin, bloodstream, and central nervous system. Within the non-neoformans Cryptococcal species, *Cryptococcal laurentii* and *Cryptococcal albidus* account for 80% of pathogenic infections. Although *C. laurentii* is found worldwide and its natural habitat remains largely unknown [17, 26], the occurrence rate 4%of its current finding is higher.

Trichosporon spp are yeast-like fungi found in soil and water. Furthermore, they belong to the normal flora of the human skin and gastrointestinal tract. The first case of onychomycosis caused by *Trichosporon mucoides* was published in 2011 by Malini A et al [27]. An additional case was published in 2015 and 2016 by Capoor et al and Rizzitelli et al [28]. The published data showed *Trichosporon mucoides* a clinical significant isolate that is consider as human pathogen even if the occurrence of this fungi rare. Currently, this fungus is only one (1/25) *Trichosporon mucoides* recovered from nail that is agreeing from previously finding. However, clinical history of this patient was not evaluated. All epidemiological papers of fungal diseases in Ethiopia were reviewed. Where there was no Ethiopia data align with this finding. Perhaps, because of these diseases are often understudied.

In the last 20 years, a change has been observed in the rates of Candida species isolated from patients with Candidiasis. The incidence of *Candida albicans* has decreased, while that of the non-albicans Candida has increased. This may be because new antifungal agents and new therapeutic strategies such as antifungal prophylaxis, secondary prophylaxis, and preventative therapy have come into use [3]. The present study is the first report including other than Candida species in Ethiopia demonstrated

as species level by this amount and variance of yeast species.

This also explains incidence of *C. albicans* has decreased, while that of the non-albicans Candida and other yeast species has increased. Although the reason for the emergence of non-albicans Candida and other than Candida species in large proportion in this study is not clear, the precision of the identification method (VITEK 2 compact system) and use of antifungal drugs for prophylaxis and treatment empirically in Ethiopia could be possible explanations.

Conclusion

Eighteen (18) different type species of yeast were isolated with *C. albicans* the predominant species in all specimen types. The emergence of non-albicans Candida and other than candida species yeast has increased. Importantly, knowledge of the species of pathogenic yeast is a useful guide to the probable pattern of susceptibility and for the successful treatment of patients. Therefore, epidemiological studies are required to determine the exact incidence and prevalence of these infections in a country's trends over time.

Availability of data and materials

The datasets used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

Acknowledgments

I would like to acknowledge the Department of Medical Laboratory Sciences, College of Health Sciences, and Addis Ababa University for giving me the opportunity to go through and develop this research. I would like to acknowledge the Ethiopian Public Health Institute Directorate of parasitic, bacterial & Zoonotic related disease unit of clinical bacteriology and mycology research case team for the primary permission to conduct this research in the bacteriology laboratory using the required equipment, supplies, and reagents Authors' contributions Elias Seyoum (ES) has participated in culture media preparation, identification of yeasts Pathogens, in drafting the manuscript, have given final approval of the version to be published; analysis and interpretation of data, and agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

Ethics approval and consent to participate

The study was conducted after it was ethically reviewed and approved by the Ethical Review board of the Department of Medical Laboratory Sciences (DRERC), School of Health Sciences, and Addis Ababa University. Written Consents were also obtained from participants. Assent form was completed and signed by family member and/or adult guardian for participants under the age of 18 years.

Competing interests, the authors declare that they have no competing interests.

Consent for publication Not applicable.

Funding No funding was received.

Reference

1. Vincent BM, Lancaster AK, Scherz-Shouval R, Whitesell L, Lindquist S. Fitness tradeoffs restrict the evolution of resistance to amphotericin B. PLoS biology. 2013;11(10): e1001692.
2. Sanguinetti M, Porta R, Sali M, La Sorda M, Pecorini G, Fadda G, Posteraro B. Evaluation of VITEK 2 and RapID yeast plus systems for yeast species identification: experience at a large clinical microbiology laboratory. Journal of clinical microbiology. 2007;45(4):1343-6.
3. Pappas PG, Kauffman CA, Andes DR, Clancy CJ, Marr KA, Ostrosky-Zeichner L, etal. Clinical Practice Guideline for the Management of Candidiasis: 2016 Update by the Infectious Disease Society of America. Clin Infect Dis. 2016;62:409-17
4. Zaoutis TE, Argon J, Chu J, Berlin JA, Walsh TJ, Feudtner C. The epidemiology and attributable outcomes of candidemia in adults and children hospitalized in the United States: a propensity analysis. Clinical infectious diseases. 2005;41(9):1232-9.
5. Malini A, Oudeacoumar P, Udayashankar C. Onychomycosis due to Trichosporon mucoides. Indian Journal of Dermatology, Venereology, and Leprology. 2011;77(1):76.
6. Yapar N. Epidemiology and risk factors for invasive candidiasis. Therapeutics and clinical risk management. 2014; 10:95.
7. Pfaller MA, Diekema DJ, Procop GW, Rinaldi MG. Multicenter comparison of the VITEK 2 antifungal susceptibility test with the CLSI broth microdilution reference method for testing amphotericin B, flucytosine, and voriconazole against Candida spp. Journal of clinical microbiology. 2007; 1;45(11):3522-8.
8. Zaragoza O, Mesa-Arango AC, Gómez-López A, Bernal-Martínez L, Rodríguez-Tudela JL, Cuenca-Estrella M. A process analysis of variables for standardization of antifungal susceptibility testing of non-fermentative yeasts. Antimicrobial agents and chemotherapy. 2011.
9. Melhem MS, Bertoletti A, Lucca HR, Silva RB, Meneghin FA, Szeszs MW. Use of the VITEK 2 system to identify and test the antifungal susceptibility of clinically relevant yeast species. Brazilian Journal of Microbiology. 2013;44(4):1257-66.
10. Donay JL, Mathieu D, Fernandes P, Pregermain C, Bruel P, Wargnier A, Casin I, Weill FX, Lagrange PH, Herrmann JL. Evaluation of the automated Phoenix system for potential routine use in the clinical microbiology laboratory. Journal of clinical microbiology. 2004;42(4):1542-.
11. Banerjee P, Haider M, Trehan V, Mishra B, Thakur A, Dogra V, Loomba P. Cryptococcus laurentii fungemia. Indian journal of medical microbiology. 2013;31(1):75.
12. Bitew A, Abebaw Y. Vulvovaginal candidiasis: species distribution of Candida and their antifungal susceptibility pattern. BMC women's health. 2018;18(1):94
13. Adjapong G, Bartlett M, Hale M, Garrill A. The isolation of Candida rugosa and Candida mesorugosa from clinical samples in Ghana. Sabouraudia. 2015;54(3):322-6.
14. Upton A, Marr KA. Emergence of opportunistic mould infections in the hematopoietic stem cell transplant patient.

Current infectious disease reports. 2006 ;8(6):434-41.
15. Mulu A, Kassu A, Anagaw B, Moges B, Gelaw A, AlemayehuM et al. Frequentdetection of 'azole' resistant Candida species among late presenting AIDS patients in north west Ethiopia BMC Infectious Diseases 2013, 13:82.
16. Sievert DM, Ricks P, Edwards JR, Schneider A, Patel J, Srinivasan A, Kallen A, Limbago B, Fridkin S. Antimicrobial-resistant pathogens associated with healthcareassociated infections summary of data reported to the National Healthcare Safety Network at the Centers for Disease Control and Prevention, 2009–2010. Infection Control & Hospital Epidemiology. 2013;34(1):1
17. Gunsilius E, Lass-Flörl C, Kähler CM, Gastl G, Petzer AL. Candida ciferrii, a new fluconazole-resistant yeast causing systemic mycosis in immunocompromised patients. Annals of hematology. 2001;80(3):178-9.
18. Liu W, Tan J, Sun J, Xu Z, Li M, Yang Q,etal.Inasive candidiasis in intensive care units in China: in vitro antifungal susceptibility in the China-SCAN study J Antimicrob Chemother. 2014; 69:162-
19. Okonkwo E C, Alo MN, Nworie O, Orji JO, Agah MV.Prevalence of oral candida albicans infection in HIVsero-positive patients in Abakaliki.American. Journal of Life Sciences. 2013; 1: 72-76.
20. Spampinato C, Leonardi D. Candida infections, causes, targets, and resistance mechanisms: traditional and alternative antifungal agents. BioMed research international. 2013;2013.
21. Colombo AL, Melo AS, Rosas RF, Salomão R, Briones M, Hollis RJ, Messer SA, Pfaller MA. Outbreak of Candida rugosa candidemia: an emerging pathogen that may be refractory to amphotericin B therapy. Diagnostic microbiology and infectious disease. 2003;46(4):253-7
22. Minces LR, Ho KS, Veldkamp PJ, Clancy CJ. Candida rugosa: a distinctive emerging cause of candidaemia. A case report and review of the literature. Scandinavian journal of infectious diseases. 2009; 41(11-12):892-7.
23. You X, Li J, Li S, Shi W. Effects of lamellar keratectomy and intrastromal injection of 0.2% fluconazole on fungal keratitis. Journal of ophthalmology. 2015;2015
24. Moges B, Bitew A, Shewaamare A. Spectrum and the in vitro antifungal susceptibility pattern of yeast isolates in Ethiopian HIV patients with oropharyngeal candidiasis. International journal of microbiology. 2016; 2016.
25. Khawcharoenporn T, Apisarnthanarak A, Mundy LM. Non-neoformans cryptococcal infections: a systematic review. Infection. 2007;35(2):51.
26. Bernal-Martinez L, Gomez-Lopez A, Castelli MV, Mesa-Arango AC, Zaragoza O, Rodriguez-Tudela JL, Cuenca-Estrella M. Susceptibility profile of clinical isolates of non-Cryptococcus neoformans/non-Cryptococcus gattii Cryptococcus species and literature review. Medical mycology. 2010;48(1):90-6
27. Smith N, Sehring M, Chambers J, Patel P. Perspectives on non-neoformans cryptococcal opportunistic infections. Journal of community hospital internal medicine perspectives. 2017;7(4):214-7.
28. Rizzitelli G, Guanziroli E, Moschin A, Sangalli R, Veraldi S. Onychomycosis caused by Trichosporon mucoides. International Journal of Infectious Diseases. 2016 ;42:61-3.