

Identification, Distribution and Antifungal Sensitivity of Candida Species in Vulvovaginal Candidiasis

Uzma Mussarat, Gohar Zaman*, Luqman Satti**, Shazia Taj, Manahil Niazi

Riphah International University, Islamabad Pakistan, *KRL Hospital, Islamabad Pakistan, **Armed Forces Institute of Pathology/
National University of Medical Sciences (NUMS) Rawalpindi Pakistan

ABSTRACT

Objective: To identify the distribution pattern and antifungal sensitivity of candida species in vulvovaginal candidiasis to improve patient safety and provide better health care.

Study Design: Descriptive cross-sectional study.

Place and Duration of Study: Department of Microbiology at the Armed Forces Institute of Pathology in association with Combined Military Hospital Rawalpindi Pakistan, from Apr 2018 to Sep 2018.

Methodology: The *Candida* strains were isolated from high vaginal swabs and were cultured on Sabouraud's dextrose agar and then stored in glycerol broth. To confirm growth, Gram staining was performed and the documentation was done using growth pattern on CHROM agar and biochemical testing with API 20 C AUX. The antifungal sensitivity pattern was checked by disc diffusion method on Sabouraud's Dextrose Agar with drug disks of Nystatin 100 units/disc, Miconazole 10µg, Fluconazole 25µg (Oxoid), Amphotericin 20µg and Clotrimazole 10µg.

Results: Among 100 collected isolates of *Candida* species 68% were *C. albicans* and 32% were non-albicans, comprising *C. glabrata* 16%, *C. tropicalis* 8%, *C. famata* 4 (4%), *C. guilliermondii* 2 (2%), *S. cerevisiae* 1 (1%) and *C. lusitaniae* 1 (1%). Moreover, *C. albicans*, *C. glabrata* (16%), and *C. tropicalis* (8%) were found to be susceptible to antifungals used in this study, while *C. famata* (4%), *C. guilliermondii* 2 (2%), and *S. cerevisiae* (1%) revealed maximum resistance against these antifungals.

Conclusion: This study display that *C. albicans* is greater than non-albicans candida in patients with VVC and non-albicans candida species are found to be almost completely resistant against commonly prescribed anti-fungal drugs.

Keywords: Antifungal sensitivity, Candida spp., Patient safety, Vulvovaginal candidiasis.

How to Cite This Article: Mussarat U, Zaman G, Satti L, Taj S, Niazi M. Identification, Distribution and Antifungal Sensitivity of Candida Species in Vulvovaginal Candidiasis. *Pak Armed Forces Med J* 2023; 73(Suppl-1): S468-473. <https://doi.org/10.51253/pafmj.v73iSUPPL-1.11857>

This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<https://creativecommons.org/licenses/by-nc/4.0/>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

INTRODUCTION

The *Candida* species reside as 10-20% of normal flora in the lower genitalia of healthy asymptomatic women and among these most important is *Candida albicans* being the root cause of 50-90% of candidiasis in humans.¹ Altered host defence system of the human body converts these commensals into pathogens. *Candida albicans* improves the immunity of the host by limiting the growth of other pathogenic fungi. "Vaginal candidiasis" or "vulvovaginal candidiasis (VVC)" is VVC is defined as inflammation of the vulvovaginal mucosa in the presence of *Candida* and absence of other infectious etiologies.²

In the last couple of decades, the spread of mycotic infections, extended hospital stay and misuse of antibiotics has led to an augmented frequency of deep fungal infections in affected populations. Statistically, it is ascertained that approximately 75%

of young women suffer from a physician-approved VVC during their lifespan^{3,4} and 5% of these infections are recurrent occurring at least 4 times in one year. Among hundred candida species, *C. albicans*, *glabrata*, *tropicalis*, *stellatoidea*, *parapsilosis*, *catemilata*, *ciferri*, *guilliermondii*, *haemulonii*, *kefyr*, and *krusei* are frequently isolated in hospital infections.⁵ In 70-90% of VVC cases, *C. albicans* is declared as the main etiological agent followed by *C. glabrata* which has been stated to reveal high prevalence and resistance to azoles.⁶

In a study carried out in tertiary care hospital in Pakistan, it was observed that vaginal candidiasis had an 18.3% prevalence in the study population.⁵ Frequently used drugs for candidiasis are amphotericin B and fluconazole, members of polyenes and azoles groups of antifungal agents respectively. However, *C. krusei* and *C. glabrata* display total resistance and decreased predisposition to fluconazole and first-line antifungal treatment with amphotericin B.⁷ This emphasized the need for precise documentation of *Candida* species and their antibiograms for successful

Correspondence: Dr Uzma Mussarat, Department of Pathology, Riphah International University, Islamabad Pakistan
Received: 08 Aug 2020, revision received: 11 Dec 2020; accepted: 20 Dec 2020

therapy of patients. In the past, *C. albicans* was considered the main etiological agent for VVC but now the involvement of non-albicans species is also evident. Therefore, knowledge of genital *candida* species and patterns of drug resistance is important in the management of patients and in preventing complications. This study is aimed to recognize different species of *candida* collected from women presenting with VVC and to detect the sensitivity pattern of collected *Candida* isolates against antifungal drugs used in this study.

In routine clinical practice, candida infections are treated with antifungal drugs without lab documentation of candida species and knowing the sensitivity pattern of the isolated *candida*. This practice is also true for a majority of hospitals in Pakistan. Deficiency of this susceptibility data of candida species to antifungal agents sometimes dilutes the effect of therapy. It has been highlighted in many studies that non-albicans *candida* like *krusei* and *glabrata* have shown diminished sensitivity to frequently used antifungals like fluconazole. On the other hand, *C. albicans* has also developed resistance against the azole group.⁸ Apropos, identification of various candida species and susceptibility patterns to commonly prescribed antifungals has become imperative for the appropriate treatment of infections caused by this organism.

METHODOLOGY

This descriptive cross-sectional data-based study was carried out from April 2018 to September 2018 at the Armed Forces Institute of Pathology (AFIP) in alliance with Combined Military Hospital Rawalpindi Pakistan. A total of 100 candida isolates were collected using high vaginal swab (HVS) specimens coming to Microbiology Labs of the Army Medical College (AMC) and AFIP, saved in glycerol broth, and later on cultured on Sabouraud's dextrose agar (SDA).

Inclusion Criteria

All pregnant, non-pregnant, and post-menopausal ladies with vaginal candidiasis were considered as inclusion criteria from the study.

Exclusion Criteria

Patients taking medication of any antifungals for any complaint and Women with infections of the reproductive tract other than vaginal candidiasis and skin disease were excluded from the study.

Considering the prevalence of vaginal candidiasis to be 18.3%⁴ in a two-year study period, the size of the sample was calculated by using the formula $N = (Z^2) Pq/E^2$ was 239. In this study, because of time and

resource limitations, the minimum desired number of samples to be analyzed was 100. The sampling technique used was Non-probability convenient sampling. Samples were collected after formal approval by the ethical review committee of Riphah International University, Islamabad (Itr no. Riphah/ERC/16/0237). All the HVS samples of candida growth were processed for confirmation and differentiation of *Candida species*.

These collected candida isolates (preserved in glycerol) were cultured on SDA plates and Gram staining was done to confirm the growth of purple-colored ovoid budding yeast and for purity of the culture. All candida isolates were inoculated onto Sabouraud's dextrose agar and the culture plate was incubated at 37°C for 48 hours, cultures were inspected for pasty, creamy, smooth, and white colonial growth of *candida*. Identification and speciation of *Candida* isolates were further done by Germ Tube Test and different color of colonies on CHROM agar culture plates. API (Analytical Profile Index) 20C AUX (Biomérieux, France) was used to perform Carbohydrate fermentation tests. The numerical profile booklet was checked for documentation of candida species with the help of manual digital entry of results. This was accomplished using the database (V4.0)* with the Analytical Profile Index: API web TM identification software - the 7-digit numerical profile was entered manually via the keyboard.

The sensitivity pattern of *candida* species against antifungals was checked on Mueller-Hinton Agar (Oxoid, UK) by disk diffusion method. Culture media was supplemented with glucose to a final concentration of 2%, which helped to provide a suitable medium for fungal growth, and the addition of 0.5 µg/ml methylene blue dye was poured to enhance the zone edge definition. The inoculated plates were kept for incubation at 35°C for 48 hours. Diameters of the Zone of inhibition were noted after 48 hours and were interpreted using approved CLSI guidelines M44-A2 Vol. 29 and the isolates were categorized as resistant, intermediate, and sensitive. The antifungal disks used were Nystatin 100 units/disc (Oxoid), Miconazole 10µg (Oxoid), Fluconazole 25µg (Oxoid), Amphotericin 20 µg (Oxoid), and Clotrimazole 10µg (Oxoid).⁹

Data analysis was accomplished utilizing Statistical Package for Social Sciences (SPSS) version 21. Each categorical variable was calculated with the assistance of Simple descriptive statistics (frequencies, percentages). The Chi-square test was pragmatic to

conclude the statistical significance and a *p*-value less than 0.05 was declared as statistically significant.

Table-I: Drug Disks and Their Zone Diameters.

Drug name	Zone Diameter, mm		
	Sensitive (S)	Dose-Dependent (DD)	Resistance (R)
Nystatin (100 units)	≥25	17-24	≤16
Clotrimazole (10µg)	≥20	12-19	≤11
Miconazole (10µg)	≥20	12-19	≤11
Fluconazole (25µg)	≥19mm	18-15	≤14
Amphotericin (20µg)	>15 mm.	-	NZ

*S: Sensitive; SDD: Sensitive Dose-Dependent; R: Resistant; NZ: No Zone

RESULTS

Gram staining of all collected *Candida* strains revealed a purple ovoid structure. Among these 68% of samples revealed the presence of germ tube while 32% of samples were found to be negative for germ tube production. Growth on CHROM agar plate indicated, 16 (16%) pinkish purple colonies that were labeled as *C.glabrata*, 8 (8%) blue colonies as *C.tropicalis*, 4 (4%) light pink colonies of *C.famata* while 68 (68%) samples displayed green colored colonies of *candida albicans*. Four samples showed blue to mauve-colored colonies which after API testing identified as *C. guilliermondii* 2 (2%), *Saccharomyces cerevisiae* 1 (1%), and *C. lusitaniae* 1 (1%). The API 20 C AUX tests recognized the *Candida* species on account of fermentation patterns and consumption of different sugars. The confirmatory test by API yielded seven species of *Candida* as a complete figure i.e. *albicans*, *C. glabrata*, *C. tropicalis*, *C. guilliermondii*, *C.famata*, *Saccharomyces cerevisiae*, and *C. lusitaniae*.

Results of Antifungal Sensitivity of C. Albicans

According to the results of growth on CHROM agar plates and API most frequently found candida species in this study was candida albicans; to be calculated as 68 (68%) of total samples. According to sensitivity results among these 68 isolated samples, 63 (93%) samples were sensitive to Miconazole, 1 (1%) samples were resistant and 4 (6%) showed dose-dependent (intermediate) response. 38 (56%) samples were sensitive to nystatin while 30 (44%) revealed dose dependent response against Nystatin drug disc. (Figure-1) Out of 68 total candida albicans, 67 (99%) were sensitive to Amphotericin and Fluconazole while 1(1%) was resistant and no sample revealed a dose-

dependent response against these two drugs. All samples were found to be susceptible to Clotrimazole.



Figure-1: Results of Antifungal Sensitivity Testing.

Comparison of Antifungal Sensitivity of All Isolated Candida Species

A comparative vulnerability pattern of isolated candida species against five antifungals used in this study has been presented in the Figure-2.

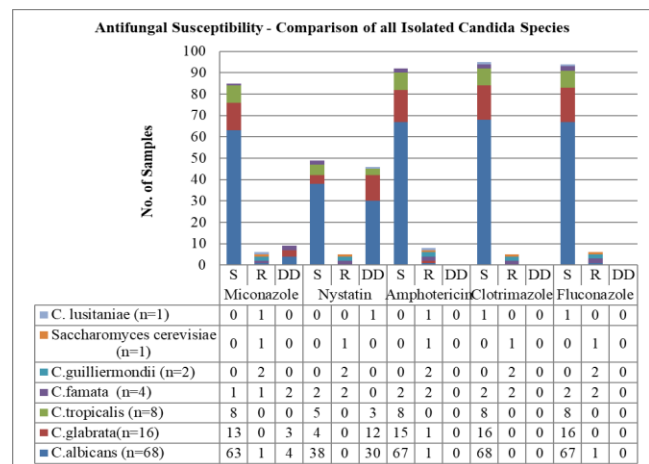


Figure-2: Comparative Analysis Sensitivity Results of All Collected Isolates.

DISCUSSION

Documentation of vaginal candidiasis relies on clinical features and direct microscopic examination but CDC recommends vaginal culture as a mandatory step before establishing the diagnosis¹⁰ Considering the different colors and consistency of colonies “CHRO Magar Candida”, provided a fast and convincing recognition of frequently found yeasts species. In this study *C. albicans* was the most frequently isolated species (68%) followed by *C. glabrata* (16%), *C. tropicalis* (8%), *C. famata* (4%), and other species (4%), these findings are similar to the study results carried out in different countries where *C. glabrata* was the second most common species isolated from

samples.^{11,12} However some studies evidence the underlining shifting trend of *Candida* infections towards NAC species.¹³

The in vitro susceptibility pattern of disease-causing candida against a particular antimicrobial drug has been evaluated to check the therapeutic response of selected drugs in the successful treatment of patients. The antifungal sensitivity pattern of fluconazole against *C. albicans* showed almost complete (99%) sensitivity and only 1% were resistant (Figure-2). This sensitivity rate is more or less equivalent to studies conducted earlier by Fuller *et al* & Brescini *et al*^{14,15} but it is contrary to the findings of study conducted by Hashemi *et al* & Adjapong *et al* which reveal lowest susceptibility against azole group.^{8,16} These differences could be due to changed geographical circumstances. Our results of antifungal sensitivity against Fluconazole are similar to the result of Ayesha *et al*, who presented *C. albicans* as highly sensitive to fluconazole exposing a resistance rate of only 1.2%, in contrast to a high level of resistance by non-albicans species of *Candida* to fluconazole.⁵ In present study all isolates of *C. guilliermondii* and *S. cerevisiae* were found to be resistant to Fluconazole. The present study shows 93% susceptibility of *Candida albicans* against Miconazole as compared to 1% resistance, 81% susceptibility was shown by *Candida glabrata* while all the *C. tropicalis* were found to be sensitive to Miconazole. These results are in accordance with the study conducted by Vanaja *et al* in Chennai, India, which showed complete sensitivity of *C. albicans*, *C. glabrata*, and *C. tropicalis* against Miconazole.¹⁷ The 50% isolated *C. famata* was sensitive to Miconazole while half were resistant. All other candida species including *C. guilliermondii*, *S. cerevisiae*, and *C. lusitaniae* showed complete resistance to Miconazole as was proven by Kanishka *et al* as well.¹⁸

According to the present study, 56% of *C. albicans* were susceptible while 44% showed a dose-dependent response against antifungal Nystatin, and no resistance was observed that is in accordance with a study conducted by Shrestha *et al* who revealed maximum susceptibility against commonly used antifungals.¹⁹ The 30% *C. albicans*, 75% *C. glabrata*, and 37% *C. tropicalis* showed dose-dependent response against Nystatin and this outcome is in accordance with the research result conducted Capoci *et al*, narrated that candida species responsible for VVC displayed the most DDS (dose-dependent sensitivity) for Nystatin^{2,20}. About 50% of *C. famata* was susceptible and 50%

were found to be resistant to Nystatin while *C. guilliermondii* and *S. cerevisiae* were resistant and *C. lusitaniae* isolates revealed a dose-dependent response.

Some studies direct that the susceptibility of *Candida* species that are linked with VVC is erratic or changeable. Regarding Amphotericin B almost all candida albicans (99%) and *C. tropicalis* were sensitive to this antifungal drug. *C. glabrata* showed 94% sensitivity and 6% resistance against Amphotericin B. These results are consistent with Lakshmi *et al*, which showed 80% susceptibility of *C. albicans* and 92% susceptibility of non-albicans candida species against Amphotericin B.²¹ The sensitivity results of Amphotericin B during this study display that this drug is sensitive against all species of *Candida* like *C. albicans*, *C. glabrata*, and *C. tropicalis*. This is consistent with a study carried out in a tertiary care hospital in Rawalpindi, Pakistan by Ayesha *et al*. A high level of resistance was seen in non-albicans candida as compared with *C. albicans* against antifungal drugs as proved by other studies.²² Moreover, 50% of *C. famata* was sensitive and 50% was found to be resistant. *C. guilliermondii*, *S. cerevisiae*, and *C. lusitaniae* showed complete resistance. Complete strains of *C. albicans*, *C. glabrata*, and *C. tropicalis* were sensitive to Clotrimazole while *C. guilliermondii* and *S. cerevisiae* strains were resistant. 50% of *C. famata* was susceptible and 50% were resistant to Amphotericin B and *C. lusitaniae* showed a dose-dependent response. In contrast to this study, Lakshmi *et al* reported 78% of *C. albicans* and 69% of non-albicans candida showed sensitivity to Clotrimazole.²¹ It has been stated that *C. lusitaniae* shows higher intrinsic resistance to amphotericin B while it is susceptible to azoles,¹⁶ while another study revealed no resistance against amphotericin B.²³

There is a difference in sensitivity patterns of *Candida* isolates in different countries that are attributable to environmental variations, the lifestyle of patients, and clinical practices that lead to altered drug response.

CONCLUSION

Regarding the overall outcomes of the present study, it has been postulated that the prevalence of *C. albicans* and susceptibility to antifungals, is still greater than in non-albicans species in patients with VVC. In addition non-albicans candida species reveal almost complete resistance against commonly prescribed anti-fungal drugs. These factors promote the recurrence of vaginal candidiasis and the persistence of infection. The evidence encourage the use of azoles for empirical therapy of uncomplicated vaginal infections due to candida species. In contrast, recurring

episodes frequently triggered by non-albicans species don't respond to these antifungals.

Fluconazole (routinely prescribed antifungal) is still an effective drug against *C. albicans* while Amphotericin B is an effective antifungal against *C. albicans*, *C. tropicalis*, and *C. glabrata* while nonalbicans candida is found to be resistant to this drug. Miconazole, Nystatin, and Clotrimazole are effective against *C. albicans*, *C. glabrata*, and *C. tropicalis* but show resistance against *C. guilliermondii*, *S. cerevisiae*, and *C. lusitaniae*.

STUDY LIMITATIONS

Data from diverse areas of Punjab and other antifungals (other than the five antifungals used in this study) could not be tested because of limited stock and availability.

RECOMMENDATION

The results of the present study can help us to predict and forecast the clinical response, which in turn can be used in an empirical selection of antifungals. This practice is recommended for a prolonged treatment plan and in a selection of alternative regimens.

Conflict of Interest: None

Author Contribution

UM: Substantial contributions to study design, acquisition of data, substantial contributions to analysis and interpretation of data, manuscript drafting and has given final approval of the version to be published.

GZ: Substantial contributions to concept, study design, critical review and has given final approval of the version to be published.

LS: Substantial contributions to analysis and interpretation of data, critical review, has given final approval of the version to be published.

ST: Substantial contributions to analysis and interpretation of data and Manuscript drafting.

MN: Substantial contributions to analysis and interpretation of data and Manuscript drafting.

REFERENCES

- Sahal G, Bilkay IS. Distribution of clinical isolates of *Candida spp.* and antifungal susceptibility of high biofilm-forming *Candida* isolates. *Revista da Sociedade Brasileira de Medicina Tropical* 2018; 51: 644-50. doi: 10.1590/0037-8682-0136-2018
- Goulart LS, Santiago EF, Ramon JL, Moura SV, Silva AR, Silva Jr IF, et al. Species distribution and antifungal susceptibility to vulvovaginal *Candida spp.* in southern Mato Grosso State, Brazil. *J Brasileiro de Patologia e Medicina Laboratorial* 2016; 52: 233-7. DOI: 10.5935/1676-2444.20170026
- Bitew A, Abebaw Y. Vulvovaginal candidiasis: species distribution of *Candida* and their antifungal susceptibility pattern. *BMC Women's Health* 2018; 18(1): 1-10. <https://doi.org/10.1186/s12905-018-0607-z>
- Rolo J, Caixeirinho P, Gomes-Ruivo P, Gaspar C, Fernandes I. Semen supports growth of *Candida albicans*: A putative risk factor for recurrence of vulvovaginal infections? *J Obstet Gynaecol Res* 2020; 46(9): 1893-9. doi:10.1111/jog.14367
- Ayesha Aslam AA, Naeem Akhtar NA, Fariha Hasan FH, Shah AA. Prevalence and in vitro antifungal susceptibility pattern of *Candida* species in a tertiary care hospital, Rawalpindi, Pakistan. 2015. doi/full/10.5555/20153189702
- Toure OA, Ama B-I, Etienne A, Henriette VB, Kady S, Berenger AA, et al. Species identification of *Candida* isolates in various clinical specimens and their antifungal susceptibility patterns in Cte d'Ivoire. *Afr J Microbiol Res* 2016; 10(2): 66-72. doi.org/10.5897/AJMR2015.7834
- Pristov K, Ghannoum M. Resistance of *Candida* to azoles and echinocandins worldwide. *Clin Microbiol Infec* 2019; 25(7): 792-8. doi.org/10.1016/j.cmi.2019.03.028
- Adjapong G, Hale M, Garrill A. A comparative investigation of azole susceptibility in *Candida* isolates from vulvovaginal candidiasis and recurrent vulvovaginal candidiasis patients in Ghana. *Med Mycol* 2017; 55(6): 686-9. doi: 10.1093/mmy/myw122
- Khan M, Ahmed J, Gul A, Ikram A, Lalani FK. Antifungal susceptibility testing of vulvovaginal *Candida* species among women attending antenatal clinic in tertiary care hospitals of Peshawar. *Infection and Drug Resistance* 2018; 447-56. doi.org/10.2147/IDR.S153116
- Al-Ahmady ZZ, Mohamed SA. Vulvovaginal candidiasis: Agents and its virulence factors. *Microbiol Res International* 2014; 2(3): 28-37.
- Felix TC, de Brito Röder DVD, dos Santos Pedroso R. Alternative and complementary therapies for vulvovaginal candidiasis. *Folia Microbiol* 2019; 64: 133-41. doi.org/10.1007/s12223-018-0652-x
- Kiasat N, Rezaei-Matehkolaei A, Mahmoudabadi AZ, Mohamadpour KH, Molavi S, Khoshayand N. Prevalence of vulvovaginal candidiasis in Ahvaz, southwest Iran: a semi-large scale study. *Jundishapur J Microbiol* 2019; 12(3): doi: 10.5812/jjm.89815
- Seyoum E, Bitew A, Mihret A. Distribution of *Candida albicans* and non-albicans *Candida* species isolated in different clinical samples and their in vitro antifungal susceptibility profile in Ethiopia. *BMC Infec Dis* 2020; 20(1): 1-9. doi.org/10.1186/s12879-020-4883-5
- Fuller J, Dingle TC, Bull A, Shokoples S, Laverdière M, Baxter MR, et al. Species distribution and antifungal susceptibility of invasive *Candida* isolates from Canadian hospitals: results of the CANWARD 2011-16 study. *J Antimicrobial Chemotherap* 2019; 74(Suppl_4): iv48-iv54. doi:10.1093/jac/dkz287
- Brescini L, Mazzanti S, Orsetti E, Morroni G, Masucci A, Pocognoli A, et al. Species distribution and antifungal susceptibilities of bloodstream *Candida* isolates: A nine-years single center survey. *J Chemotherap* 2020; 32(5): 244-50. doi.org/10.1080/1120009X.2020.1783154
- Hashemi SE, Shokohi T, Abastabar M, Aslani N, Ghadamzadeh M, Haghani I. Species distribution and susceptibility profiles of *Candida* species isolated from vulvovaginal candidiasis, emergence of *C. lusitaniae*. *Current Med Mycol* 2019; 5(4): 26. DOI: 10.18502/cmm.5.4.2062
- Vanaja R, Sudha V, Adishes M. Isolation and Sensitivity Pattern of Genital *Candida* Species in STD OP Female Attendees of Tertiary Care Hospital, Chennai, India. *Int J Curr Microbiol App Sci* 2016; 5(8): 525-8.
- Das KH, Mangayarkarasi V, Sen M. Antifungal resistant in non-albicans *Candida* species are emerging as a threat to antenatal women with vulvovaginal candidiasis. *Biomedical and Pharmacol J* 2019; 12(2): 1369-78. doi.org/10.13005/bpj/1765

Vulvovaginal Candidiasis

19. Shrestha P, Pokharel SM, Shrestha A. Antifungal susceptibility pattern of candida isolates causing vulvovaginitis in reproductive age women. *Tribhuvan Uni J Microbiol* 2020; 7: 1-7. doi.org/10.3126/tujm.v7i0.33784
 20. Capoci IRG, Bonfim-Mendonça PdS, Arita GS, Pereira RRdA, Consolaro MEL, Bruschi ML, et al. Propolis is an efficient fungicide and inhibitor of biofilm production by vaginal *Candida albicans*. *Evidence-Based Complementary and Alternative Medicine* 2015; 2015. doi.org/10.1155/2015/287693
 21. Lakshmi N, Kumari GR, Purushotham MD, Krishna PBM. Isolation and Speciation of *Candida* from Vulvovaginitis and their Antifungal Susceptibility. *Int J Curr Microbiol App Sci* 2015; 4(12): 121-9.
 22. Singh M, Chakraborty A. Antifungal drug resistance among *Candida albicans* and Non-*albicans candida* species isolates from a tertiary care centre at Allahabad. *J Antimicrob Agents* 2017; 3(150): 2472-1212. DOI: 10.4172/2472-1212.1000150
 23. Khan Z, Ahmad S, Al-Sweih N, Khan S, Joseph L. *Candida lusitanae* in Kuwait: Prevalence, antifungal susceptibility and role in neonatal fungemia. *PLoS One* 2019; 14(3): e0213532. doi.org/10.1371/journal.pone.0213532
-