A Tale of Candidal Infections from a Tertiary Care Centre

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Abstract: This study was aimed to probe the prevalence of Candida species in a tertiary care hospital, Kashmir, India. 4200 samples were directed to the microbiology lab for evaluation including urine(47%), blood(23.8%), sputum(7.9%), pus(4.7%), tracheal fluid(4.7%), pleural fluid(3.3%), CSF(2.6%), vaginal swab(0.7%), BAL (0.64%) and oral swab(0.11%). Candida species were isolated from 151 clinical samples of which Candida species were isolated from 36(23.8%) immunocompromised patient samples. Out of 151 positive samples, Candia species was mainly isolated from urine samples (54.3%) followed by sputum (26.6%) and vaginal swabs (7%). Candida species isolated from 151 samples showing growth of Candida included Candida albicans 101 (66.8%) and non-albicans Candida species 50 (33.1%). Among non-albicans Candida species Candida parapsilosis (36%) was the commonest isolated followed by Candida glabrata(26%) and Candida tropicalis(16%). isolated followed by For the isolated of Candida species Sabouraud dextrose agar with Chloramphenicol was used and for the identification of different Candida species HiCrome agar, germ tube test, Morphology on corn meal agar, Carbohydrate assimilation tests were performed. <u>Conclusion</u>: overall prevalence of Candida species was noted to be (3.5%). Predominant species was Candida albicans (67%) isolated among the patients attending the tertiary care hospital, Kashmir, over 5 months of period.

Keywords: Candida albicans, non-albicans Candida species, HiCrome Agar, Germ tube, Corn Meal Agar.

1. Introduction

Fungal infections have considerably increased which is a cause of mortality. Successive use of immunonosuppressants and antibiotics in immunocompressed persons is a major cause of these infections. [1]Pathogenicity caused by Candida is most frequently found in immunocompetent as well as in immunocompromised persons. [2]Different species belonging to genus Candida are the causative agents of endogenous infections e.g.; Candida albicans responsible for various fungal infections and are usually found colonized in the skin and mucosal surfaces of humans. [3] As per the recent studies there has been tremendous increase in fungal Non-albicans infections due to Candida in or immunocompromised patients which becomes lifethreatening.[4] Antibiotic resistance is seen in Candida species especially to azoles due the expression of the efflux pumps which decrease the drug sensitivity and alter the structure of antifungal target proteins.[5]

2. Material and Methods

The study was performed in the department of Microbiology at Sheri Kashmir Institute Of Medical Sciences (SKIMS), Kashmir, a tertiary care hospital catering the state population.

Samples accepted from different wards and OPDS were processed over 5 months of period from January 2018- May 2018.

Specimen collection and processing

The various clinical specimens were collected under aseptic conditions in a sterile leak proof container accompanied by a relevant clinical history and rapidly transported to the laboratory and processed as per the standard microbiological procedures. **Direct Microscopy;** Fluid specimens such as urine, sputum, Cerebrospinal fluid (CSF) etc were used to make wet mounts or smears for the detection of budding yeast cells, pseudohyphae and/ or hyphae.

Culture; For the isolation of *Candida* species specimens were transferred onto appropriate fungal culture media i.e Sabouraud Dextrose agar with Chloramphenicol and incubated at 37^{0} C.

Growth on HiCrome agar; For the identification of *Candida species* Chromogenic media was used to isolate and identify clinically important *Candida species*, particularly *Candida albicans*, *Candida tropicalis*, *Candida glabrata* etc based on colored colony morphology depending upon the reactions between the specific enzymes of the different species and the Chromogenic substances. The pure culture was seeded onto the HiCrome agar media and incubated at 37°C for 48 hours. The media was observed for characteristic color change as per HiMedia technical data. [6]

Germ tube test; This was the rapid test for presumptive identification of *Candida albicans*. Pure colony of yeast was suspended in the eppendorf containing human serum and incubated at 37° C for 2-3 hours. Filamentous extension from yeast cell was observed. [7] (figure1)

Morphology on Corn Meal agar; Nutritionally deficient media that provides excellent medium for yeasts to form pseudohyphae. Isolated colonies were inoculated on corn meal agar over which a cover slip was laid and incubated at 37^{0} C for 48 hours, and examined under microscope for pattern of blastoconidia and pseudohyphae and presence of Chlamydospores in case of *Candida albicans*. (Figure2)

Carbohydrate assimilation; Identification of Candida species is performed by checking the ability of yeast to utilize a specific carbohydrate as sole carbon in presence of

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oxygen. Different 11 sugar discs used for this test were dextrose, sucrose, maltose, lactose, galactose, cellobiose, melibiose, inositol, dulcitol, xylose, and trehalose. All these discs were placed on Yeast nitrogen base agar with Inoculum and the plate was incubated at 37^{0} C for 48 hours and the plates were observed for zone formation around the discs. [8] (figure3)

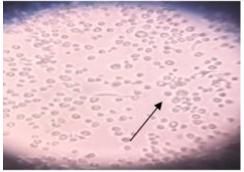


Figure 1: Germ tube test Germ tube test in *C.albicans*

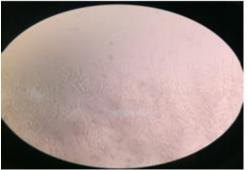


Figure 2: Morphology on CMA Morphology of *C.parapsilosis*

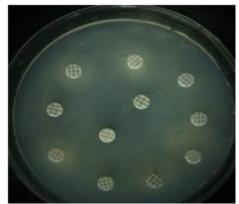


Figure 3: Carbohydrate assimilation test Carbohydrate assimilation of *C.kefyr*

3. Observations and Results

From January 2018- May 2018, Out of 4200 samples sent to the microbiology lab for evaluation, urine were 2000(47%), blood 1000(23.8%), sputum 333(7.9%), tracheal fluid 200(4.7%), pus 200(4.7%), pleural fluid 140(3.3%), CSF 113(2.6%), vaginal swab 31(0.7%), BAL 27(0.64%) and Oral swab 5(0.11%) in number. Of the 151 patients from whom Candida spp. were isolated 34 (22.5%) were immunocompromised which included 8 patients with diabetes, 3 patients with Kidney transplant, 21 patients with different malignancies and 2 patients with connective tissue disorder. Candida spp. were isolated from 151 clinical samples, out of 151 positive samples , urine samples positive for Candida spp. were 82 (54.3%).Blood samples showing growth of *Candida spp.* were 6(3.9%).40(26.6%) of Candida spp. were isolated from sputum. Candida spp. positive for tracheal fluid was 3 (1.9%). Pus samples positive for Candida spp. were 3(1.9%). Pleural fluid positive for fungus were 3(1.9%). CSF samples positive for fungus were 1(0.0.66%). The Candida spp. isolated from vaginal swabs were 11(7.2%). The Candida spp. isolated from BAL samples were 1 (0.66%). Oral swab positive for fungus were 1(0.66%) which was Candida albicans. (Figure **4**)

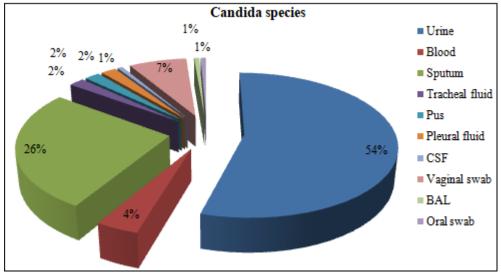


Figure 4: Showing distribution of samples positive for Candida spp.

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Among the 82 *Candida spp.* isolated *from* urine *Candida albicans* were found in 47(57%) and non-albicans Candida species in 35(43%). Of the 35 non-albicans Candida, *Candida parapsilosis* 12(34%), *Candida glabrata* 9(25.7%), *Candida tropicalis* 6 (17.1%), *Candida rugosa* 2(5.7%), *Candida krusei* 2 (5.7%), *Candida spherica* 1(2.8%), *Candida auris* (2.8%), *Candida lusitaniae* (2.8%), *Candida famata* (2.8%), *Candida dubliniensis* (2.8%) were isolated.

Of the 6 blood samples positive for *Candida spp. Candida albicans* were seen in 2(33%) cases and non-albicans Candida in 4(66%).All the non-albicans Candida were *Candida parapsilosis* (100%).

Of the 40 sputum samples positive for *Candida spp*, *Candida albicans* were isolated in 37 patients (92.5%). The 3 (7.5%) non-albicans were C. tropicalis (66%), *Candida kefyr* (33%).

Among the 3 positive pus samples showing the growth of *Candida spp, C. albicans* was seen in 2(66%) and non-albicans in 1(33%) which was *Candida parapsilosis* (100%).

Of the 3 pleural fluid samples positive for *Candida spp*, *Candida albicans* was seen in 1 (33%) and non *albicans* Candida in 2 (66%) cases, including *Candida krusei* 1 (50%) and *Candida famata* 1 (50%).

Of the 1 positive CSF samples non- albicans Candida was isolated and the isolated non-albicans *Candida* seen was *Candida parapsilosis* 1 (100%).

Among the 11 vaginal swab samples which were showing the growth of *Candida spp*, predominantly isolated spp. was *Candida albicans* in 8(72%) and non-albicans in 3(28%) cases. All the non-albicans *Candida* spp. was *Candida glabrata* (100%).

The *Candida* species isolated from 1 positive BAL sample was non-albicans Candida, including *C.kefyr* 1 (100%) Of the 1 Oral swab positive for *Candida* spp, the isolated species was *C.albicans* 1(20%) (**Figure5&6**)

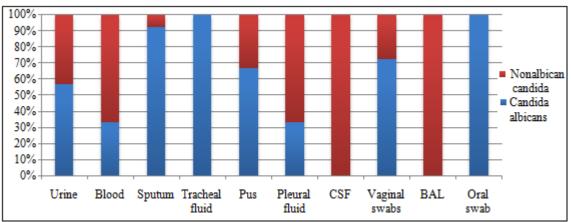


Figure 5: Comparison of Candida albicans and non-albicans Candida species from different clinical specimens

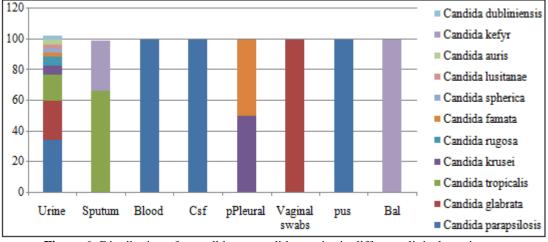


Figure 6: Distribution of non-albicans candida species in different clinical specimens

So out of total 4200 samples sent to the lab, 151(3.5%) samples were positive for *Candida* spp. Among them *Candida albicans* was predominant seen in 101(66.8%) cases and non albicans Candida in 50(33.11%).

4. Discussion

The present study was carried out to know the prevalence of *Candida species* in a tertiary care hospital and it was concluded that the incidence of infections from *Candida albicans* was more predominant than non-albicans Candida

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spp. among which *Candida parapsilosis* was the most common isolated species.

While comparing my study with Kee Peng Ng *et al.*, (2013) ^[9] on surveillance of *Candida spp*. the authors found that *Candida albicans* was predominant *Candida spp*. isolated from various clinical samples which are in conformity to the present studies. Similarly Shivanand *et al.*, (2011) ^[10] study was in conformity to ours. In contrast, a study by Ravinder Kaur *et al.*, (2015) ^[11]on epidemiology of Candidiasis and antifungal susceptibility, found the incidence of Candidiasis caused by non-albicans Candida spp. (63.3%) was higher than *Candida albicans* (36.7%).

Candidemia was caused due to non-albicans Candida followed by *Candida albicans*. These results are different than those seen by Vicky Gandhi., $(2017)^{[12]}$, where they found *C. albicans* was responsible for Candidemia.

In the present study most commonly isolated species among non-albicans Candida was *Candida parapsilosis* in the neonates which are similar to the studies conducted by Wilson *et el.*, $(2015)^{[13]}$.Among the *Candida spp.*, the most common species isolated were *Candida parapsilosis* (36%), *Candida glabrata* (24%), *Candida tropicalis* (16%).Whereas, a study conducted by *Manisha et al.*, $(2016)^{[14]}$, found that the most common species isolated were Candida tropicalis (62.25%) followed by *C.glabrata* (23.84%).

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