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SPECIES DISTRIBUTION OF CANDIDA ISOLATES FROM PATIENTS, TREATED AT UMBAL-STARA ZAGORA

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ABSTRACT

This study presents data on species distribution of Candida species in clinical specimens from University Hospital in Stara Zagora for three years period. The 18,947 samples were examined and 522 strains were isolated. Causative agents of candida infection were found most often in the lower respiratory tract (36.40 %), in the upper respiratory tract (22%), in the urogenital system (12%) and others. A large number of isolates were assigned to the genus Candida dominated by Candida albicans (61%), Candida glabrata (11%), Candida krusei (11%), Candida tropicalis (9%). The isolates from invasive mycoses were 0.95 % of all Candida isolates.

Key words: Candida albicans, Candida glabrata, Candida krusei, Candida tropicalis

INTRODUCTION

Fungal infections have increased enormously in recent decades, often as a result of advanced medical treatment and the increase in the number of immunocompromised patients. Candida albicans is still the most frequent cause of fungal infections. However, the use of broad-spectrum antibiotics and antifungal agents for prophylaxis has led to a shift in the epidemiology and etiology of Candida and non-Candida yeast species infections (1, 2). Although Candida albicans has a leading role, more commonly reported cases of Candida non-albicans isolates are reported (3).

Numerous factors have contributed to the increase in fungal infections. The most important is an ever-expanding immunocompromised population with mucosal or cutaneous barrier disruption, defects in the number and function of neutrophils or in cell-mediated immunity, metabolic dysfunction, and extremes of age (4).The increased use of broad spectrum antibiotics, cytotoxic chemotherapy and transplanted central venous catheters, surgical infections increase the risk of Candida infections (5). The use of triazole antifungal agents led to the selection and increasing incidence of colonization or infection with less susceptible or resistant non-albicans strains, such as Candida glabrata, Candida krusei, Candida tropicalis, and Candida dubliniensis. Candida spp. are fourth in frequency as a cause of nosocomial infection. (5, 6, 7).

This data analyses species distribution of Candida isolates in clinical specimens tested for 3 years period (2009 -2011).

MATERIALS AND METHODS

The Laboratory of Clinical Microbiology, Stara Zagora performed microbiological diagnosis of patients treated at University Hospital. Specimens were tested for bacterial pathogens and agents causing fungal infections. Two basic approaches were applied: microscopic methods and culture facilities followed by subsequent identification. The cultivation was done on blood agar and Sabouraud dextrosa agar, production of Becton Dickinson and NCZPB. Laboratory methods for identification were based on a study of various phenotypic characteristics, including colony morphology and microscopic observation, assimilation of sugars and fermentative reactions. We used a variety of different commercial identification systems

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based on the color change due to the use of certain substrates for yeast metabolism.

We used commercial non-automated methods- Fungifast (EliTech France) and Integral System Yeast Plus, API 20 AUX (bioMerieux) and the following morphological tests: 1.Corn meal agar-Tween 80 stimulates sporulation of C. albicans, and is useful in suppressing certain other fungal growth. Chlamydospore production is an important diagnostic characteristic used in the identification of C. albicans; 2.Chrom agar- Candida is a selective medium for the isolation and presumptive identification of yeast and filamentous fungi and differentiation of C. albicans, C. tropicalis, C. krusei and C.glabrata;3.Germ tube test- a germ tube test is a diagnostic test in which a sample of fungal spores are suspended in serum and examined by microscopy for the detection of any germ tubes. Positive germ tube test is strongly indicative for Candida albicans.

Study of the sensitivity to antifungal agents was performed on Candida isolates from system mycoses.

RESULTS AND DISCUSSION

During the analyzed period, the 18 947 clinical specimens were studied; the distribution of specimens is presented in **Table 1.**

Table1. *Distribution of specimens*

Total number of samples of types of materials for the period 2009 - 2011				
materials	2009 6890	2010 4800	2011 7257	Total 18947
blood culture	475	502	761	1738(9, 17%)
wound secretions	392	450	273	1115(5, 88%)
urine	1922	1298	2175	5395(28, 47%)
specimens from the upper respiratory tract	2837	1394	2712	6943(36, 64%)
specimens from the lower respiratory tract	319	565	715	1599(8, 44%)
cerebrospinal fluid	381	110	143	634(3, 35%)
specimens from urogenital tract	66	161	185	412(2, 17%)
fecal specimens	423	211	168	802(4, 23%)
others	75	109	125	309(1, 63%)

Samples of the upper respiratory tract represented 36.64% of the total samples, followed by urine, blood cultures, lower respiratory tract, wound secretions, fecal specimens and others. From these samples were isolated totally 522 strains of fungi, all belonging to the genus Candida. The 320 of them were identified as C. albicans, 59 were C.glabrata, 55 were C.krusei, 47 were C. glabrata, 47 were C. tropicalis, and 21 were C. non albicans

Analysis and distribution of species in different materials are presented in **Table 2.** Most frequently were Candida species of lower respiratory tract- 36.40% of totally positive samples, dominated again by Candida albicans (64, 73%). Materials were initially evaluated by direct microscope examination for presence of cellular elements to rejected samples from oral cavity and if it is necessary the study is repeated. Isolates from the respiratory tract represented 22% of totally isolated Candida. At the highest rate- 72% were identified as Candida albicans. Many of nasopharyngeal secretions and samples from the mouth came from patients with systemic blood diseases, some of them with neutropenia. There were a large number of isolates from the urogenital materials (12%), mainly in women with acute and recurrent vulvovaginal candidiasis, but also in other diseases of the genital tract. Rarely fungi were isolated from men. From 5395 studied urines, the 55 isolates were Candida spp. with a predominance of C.

albicans. The determination of candiduria as systemic candidiasis requires a combination with other disease manifestations. Candida isolates were found in 1% of the studied materials in surgical wounds and infections, including skin infections, in patients with severe surgical diseases such as peritonitis, deep wound infection and others were frequently observed invasive mycoses. The proof of invasive mycosis is a challenge for any laboratory considering the severity of clinical signs and the danger for the life of patients. There were 5 (0.95%) proven cases of disseminated fungal infections. The data obtained were lower than many data that have been reported (7, 8). Probably a long period of cultivation under aeration will increase that percentage, considering the large number of uncertain temperature and severe conditions without determination of the cause. Clinical cases without proven etiological agent require the use of other methods for diagnosis of fungi, such as determination of antigens and antibodies, immunofluorescence techniques, automated identification systems and etc. It is necessary to examine the antifungal susceptibility because of the increasing resistance of Candida isolates.

Table2. *Analysis and distribution of species in different materials*

Candida species from different clinical specimens at the University Hospital - Stara Zagora for the period 2009 - 2011						
species/materials	C.albicans	C.tropicalis	C.glabrata	C.krusei	C.non-albicans	total
blood culture	2			2	1	5
wound secretions	6	3	1	3		13
urine	28	7	10	8	2	55
specimens from the upper respiratory tract	83	12	8	10	2	115
specimens from the lower respiratory tract	123	18	20	19	10	190
cerebrospinal fluid	1	1	3		2	7
specimens from urogenital tract	40	2	13	4	4	63
fecal specimens	22	3	9	6	7	49
others	15	1	4	3	2	25
total	320	47	59	55	21	522

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