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Original scientific paper

The Importance of Serological Tests Implementation in Disseminated Candidiasis Diagnose

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A B S T R A C T

Candidiasis is defined as an infection or disease caused by a fungus of the genus Candida. Rate of disseminated candidiasis increases with the growth of the number of immunocompromised patients. In the the last few decades the incidence of disseminated candidiasis is in growth as well as the mortality rate. The aim of this survey is to show the importance of serological tests implemenation in disseminated candidiasis diagnose. This is a prospective study involving 60 patients with malign diseases with and without clinical signs of disseminated candidiasis and 30 healthy people who represent the control group. Apart from hemoculture, detection of circulating mannan antigen and adequate antibodies of Candida species applying comercial ELISA test was determined in each patient. This survey deals with relevant factors causing disseminated candidiasis. This survey showed that the group of patients with clinical signs of disseminated canididiasis had more patients with positive hemoculture to Candida species, then the group of patients without clinical signs of disseminated candidiasis. The number of patients being examined and positive to antigens and antibodies was higher ($p<0.01$) in the group of patients with clinical signs of disseminated candidiasis (7/30; 23.3%), then in the group of patients without clinical signs of disseminated candidiasis (0/30; 0%). Average value of titra antigen was statistically higher ($p<0.001$) in patients with Candida spp. positive hemocultures rather then in patients with Candida spp. negative hemocultures. In the group of patients with clinical signs of disseminated candidiasis 6/30 (20%) of patients had Candida spp.positive hemocultures while in the group of patients without clinical signs of disseminated candidiasis 1/30 (3.3%) of patients had Candida spp. positive hemocultures, which was considerably higher ($p<0.05$). Correlation of results of hemoculture and mannan antigens and antibodies in patients with disseminated candidiasis were statistically significant, while correlation of results of hemoculture and antibodies was insignificant. Because of low sensitivity of hemoculture and time needed for isolation of Candida spp., introducing serological tests in regular procedures would speed disseminated candidiasis diagnose.

***Key words**: Candida, mannan antigen, disseminated candidiasis*

Introduction

Disseminated candidiasis is defined as multiorganic fungus infection caused by species of genus *Candida* with or without candidemia[1]. This infection usually involves brain, kidneys, heart, eyes and other organs and systems. The rate of disseminated candidiasis grows with the increase of immunocompromised patients and it is also connected with invasive interventions especially in departments of intensive care unit. Yeasts are major cause of infections and they take the fourth place among microorganisms which are isolated in hemocultres (4.9%)[2]. In the last few decades the incidence of disseminated candidiasis is noticed and it takes 8 cases *per* 100000 of the population in the USA. According to Centre of Disease Control (CDC) and Departments of Bacteria and Fungus Infections data the rate of mortality is 30–50%. Gastrointestinal tract (0.55%), vagina (2.2–68%), and skin (approximately 2%) represent the large reservoir of *Candida albicans* (*C. albicans*). After impairing the mu-

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cosa of gastrointestinal tract *Candida* can expand to different organs leading to disseminated infection. Patients with hematological impairment, patients with impairment of organs, patients who had bone marrow transplantation, patients who had operated gastrointestinal tract or heart, patients with hard burns, patients with prolonged intravein catheterization, and patients with diabetes mellitus are prone to disseminated candidiasis[3]. The risk factors for Disseminated candidiasis are neutropenia and impaired mucosa as a consequence of chemotherapy or X-rays as well as prolonged use of antibiotics[4].

Clinical signs of disseminated candidiasis are unspecific. The most common clinic signs are fever, sepsis, changes on the skin and retina. These symptoms can be found in other diseases as well which makes it harder to diagnose disseminated candididasis. Fever and sepsis do not react to therapy of large spectrum antibiotics and they are very often the only clinical signs of acute disseminated candidiasis. Making an early diagnosis of disseminated candidiasis is very hard, since the clinical signs and symptoms are very unspecific which postpones beginning of antifungal therapy. The classic diagnose of disseminated candidiasis is based on isolation of *Candida* species (*Candida* spp.) from blood and on histopathological and microbiological analyses of the bioptical material. Disadvantages of these diagnosing prcedures are evident in low sensitivity of ones while the others are very invasive and often impossible to do in practice[5].

Apart from these methods serology methods have developed lately and they are based on detection of antibodies and antigens of *Candida* from body fluids. They represent the early specific markers of disseminated infection[6]. The basic advanatge of serology method in comparison to classic labaratory method is that the result is given within few hours with specificity of 93% and sensitivity of 80%, while hemoculture becomes positive in 2 to 5 days an it remains negative in 50% of cases[7]. Antibodies and mannan antigen detection tests can be made with double immunodiffusion (ID) and enzyme immunoassay (ELISA-Platelia *Candida* antibody test). Mannan antigen detection is performed by several techniques like radioimmunoassay (RIA), enzyme imunnoassay (ELISA), latex agglutination (LA) and reserve passive latex agglutination (RPAL). ELISA test is based on detection of α–oligo mannose from serum which is released from cell walls during *Candida* infection[8]. Combination of these two test, detection of antibodies and antigens, leads to higher specificity and sensitivit in early disseminated candidiasis diagnose as well as early usage of antifungal therapy[9].

Aims of Survey

1) To define serum values of manan antigen and antibodies in patients with clinical signs of disseminated candidiasis.

2) To correlate the results of hemoculture with the results of specific antigen of *Candida* spp. (mannan antigen) and antibodies in patients with clinical signs of disseminated candidiasis.

Materials and Methods

This survey was made at Clinic for Laboratory Diagnosis, Department of Microbiology, University Clinic Centre in Tuzla. This survey includes 90 patients. Test group I was made of 30 immunocompromissed patients (patients with malign diseases who underwent cytostatic therapy) with clinical signs of disseminated candidiasis. Test group II was made of 30 immunocompromissed patients (patients with malign diseases who underwent cytostatic therapy) without clinical signs of disseminated candidiasis. Clinical signs which included patients in test group I were:

- neutropenia $<1.5 \times 10^9$/L lasting longer than 10 days
- fever-lasting more than 96 hours without any response to antibiotic therapy
- body tempertaure higher than 38 °C.

Control group was made of 30 healthy adults (blood donors) whose samples were recived from Department of Transfusiology from University Clinic Centre in Tuzla. From histories of diseases of patients personal data was taken (name, surname, date of birth and gender/sex). Apart from this data, relevant data for making disseminated candidiasis diagnose like information about disease, information about any antibiotic therapy, information about values of neutrophil leucocytes and information about body temperature was also taken. To perform microbiological examination 10 ml of blood for hemoculture and 3 ml of blood for serology testing was taken from each patient. In original basis for hemoculture BD Bactec Mycosis IC/F 10 mL of blood was put and cultivated for 14 days on 30 °C in the machine for automatic detection Bactec 9120 (Becton, Dickinson and Company). After the signal which marks the increase of yeasts in hemoculture the same were disseminated to SDA, Malt Agar an incubated for 48 hours on 27 °C. Identification of increased colonies of *Candida* was made by standard mycological procedures (microscope, germination test and chlamydospore test). Germ tube test was made by incubation of increased colonies of yeasts from nutrient foundation (optimal inoculum 104–16 cells *per* mL), on 37 °C in 10% human serum for two hours. The result of blastospor with formed cylinder shoots which were 8–10 μm long and 2–3 μm wide was considered as a positive test. Proving the ability of making hlamidospore was done by cultivating increased colonies of yeasts of potato agar, which was incubated 3–5 days on 27 °C in a humid chamber. The result of characteristic terminal, intercalar and lateral hlamidospore on pseudohyphae of yeasts was marked as positive test. Apart from standard mycrobiological methods for detection of yeasts of *Candida* species automatic system for identification of microorganisms was also used applying VITEK 2 Compact apparatus. The presence of antibodies in mannan antigen C.

species was determined by applying ELISA Platelia *Candida*-specific Ab test (Bio-Rad Laboratories) and the presence of mannan antigen was determined by applying ELISA Platelia *Candida*-specific Ag test (Bio-Rad Laboratories). Performing of the tests was made by instructions of manufacturer[8]. All samples with concentration of mannnan antigen lower than 0.25 ng/mL (C<0.25) were marked as negative. If the concentration of antigen was betweeen 0.25 and 0.5 ng/mL (0.25≤C<0.5) the tests were marked as intermediary. Concentration of mannan antigen higher or equal to 0.5 ng/mL (C≥0.5) was marked as positive. All samples of serum with concentration of anti-mannan At lower than 5 AU/mL (C<5) were considered negative. Samples of serum with concentration of anti-mannan At between 5 and 10 AU/mL (5≤C<10) were considerd as intermediary. Samples of serum with concentration of anti-mannan At higher or equal to 10 AU/mL (C≥10) were considred as positive.

Statistical data treatment

In statistical data treatment standard method of descriptive statistics (correlation of values) was used. For testing statistical significance between the groups parametric and unparametric tests, student's t-test, Spearman's coefficient of correlation and ANOVA were used. Statistical hypothesis was tested on p=0.05 level, i.e. the difference between the groups was considered as important if p<0.05.

Results

From the total of 90 patients 30 belong to control group. From the remaining 60 patients, 37 (61%) of them were with hematological and 23 (39%) of them were with oncological types of malignity. Presence of patients in both groups according to the type of malignity was 7/60 of patients with double antifungal therapy and 11/60 patients with at least one antifungal in their therapy. 42/60 of patients did not have antifungal s included in their therapies. In the group of patients who did not have antifungal s in their therapy 5/42 (11.9%) was with positive hemoculture. In the group of patients who had antifungal s in their therapy 2/18 (11.1%) was with positive hemoculture. This difference is not statistically significant. More (p<0.001) patients with positive antigen was found among the patients who did not have antifungal therapy (10/15; 66.7%) in comparison to patients who had antifungal therapy (3/45; 6.7%) (Figure 1). There was not any significant difference between the presence of antibodies in the group of patients who had (23/45; 51.1%) and in the group of patients who did not have (9/15; 60%) antifungal therapy (p>0.76).

In both groups 2/60 of patients had 4 antibiotics in their therapy, 13/60 of patients had triple AB therapy, 15/60 of patients had double therapy nad 11/60 of patients had at least one antibiotic in their therapy. 19 of patients did not have any antibiotics in their therapy. In the group of patients with clinical signs of disseminated candidiasis all patients had antibiotic therapy 30/30. In

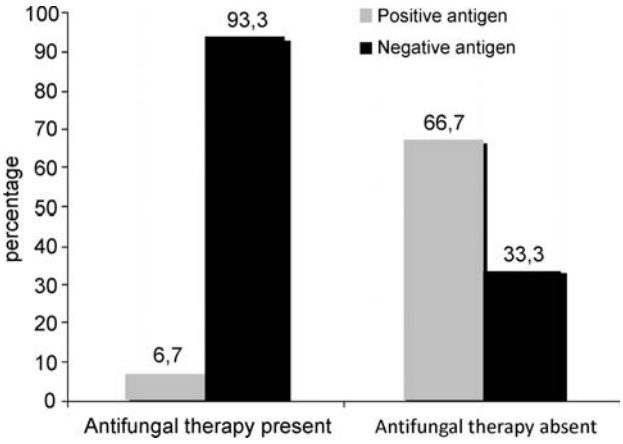

Fig. 1. The correlation of applied antifungal therapy and the presence of mannan antigen.

the group of patients without clinical signs of disseminated candidiasis 11/30 (36.6%) of patients had antibiotic therapy which was statistically significant.

Values of body temperature

Average body temperature in the group of patients with clinical signs of disseminated candidiasis was 38.8± 0.4 °C, while the average body temperature in the group of patients without clinical signs of disseminated candidiasis was 37.6±0.8 °C. It was approximately higher for 1.19 degrees (%95CI = 0.9–1.5), which was statistically significant difference (p<0.001). The body temperature of all patients in the control group was normal.

Values of neutrophil leucocytes

The values of neutrophil leucocytes were lower (p< 0.001) in patients with clinical signs of disseminated candidiasis (0.69±0.36), in comparison to patients without clinical signs of disseminated candidiasis (2.82± 2.27), with approximate difference of 2.13 (%95CI = 1.29–2.97). The control group had normal values of neutrophil leucocytes (Figure 2). 13/30 (43.3%) of patients had the values of neutrophil leucocytes $<0.5 \times 10^9$/L. From these in 2/13 (15.3%) of patients disseminated candidiasis was proved.

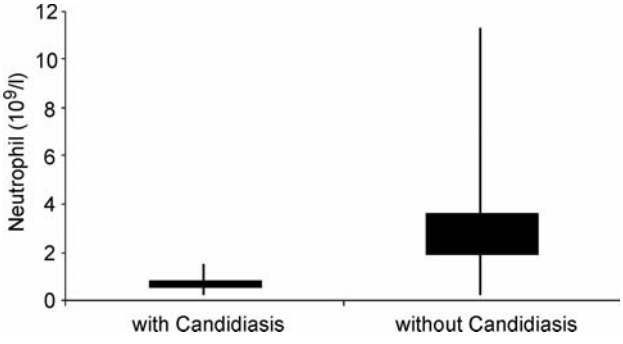

Fig. 2. Relation of values of neutrophil leucocytes in patients with and without clinical signs of disseminated candidiasis.

Titra antibodies and antigen values analysis with the results of hemoculture

In the group of patients with clinical signs of disseminated candidiasis 6/30 (20%) of patients had positive hemoculture to *Candida.* In the group of patients without clinical signs of disseminated candidiasis 1/30 (3.3%) of patients had positive hemoculture to *Candida*, which was statistically higher ($p<0.05$).

Average value of titra antibodies in patients who had positive hemoculture to *Candida* was 20.7±16.1 AU/mL while average value of titra antibodies in patients who had negative hemoculture to *Candida* was 13.7±15.5 AU/mL. This was not statistically significant ($p=0.94$), as shown in Figure 3. The average value of titra antigen

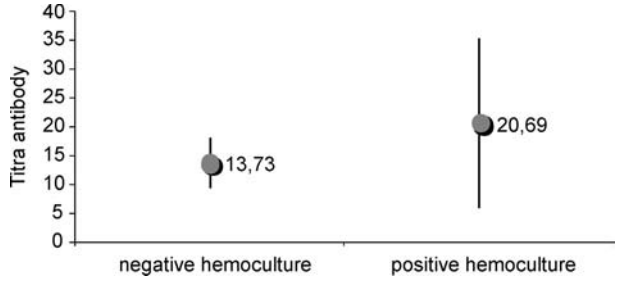

Fig. 3. Relation of values titra antibodies and hemoculture.

(0.86±1.05) ng/mL was statistically higher in patients with positive hemoculture ($p<0.001$), in comparison to patients with negative hemoculture (0.13±0.45) ng/mL, with approximate difference of 0.73 (%95CI = 0.30–1.17). This difference is shown in Figure 4. The average value of titra antigen in the control group was 0, while the average value of titra antibodies was 0.02. This number is lower in the comparison to the other two examined groups ($p<0.05$).

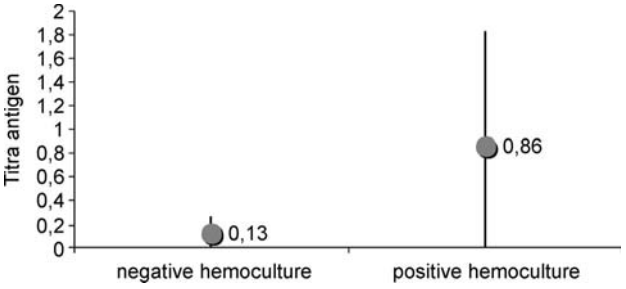

Fig. 4. Relation of values titra antigen and hemoculture.

Average values of titra antigen and titra antibodies in patients

The average values of mannan antigen in the group of patients with clinical signs of disseminated candidiasis was 0.43±0.78, which was statistically more ($p<0.05$) than in the group of patients without clinical signs of disseminated candidiasis as well as in the control group, where there were not any patients with detectable values of this antigen. The average values of antibodies were higher ($p<0.05$) in the group of patients with clinical signs of disseminated candidiasis (15.85±16.24), in comparison to the group of patients without clinical signs of disseminated candidiasis (13.24±15.0), and in comparison to the control group (0.01±0.06). The number of patients who were positive to antigen and antibodies was higher ($p<0.01$) in the group of patients with clinical signs of disseminated candidiasis (7/30; 23.3%), than in the group of patients without clinical signs of disseminated candidiasis (0/30; 0%) (Figure 5). The number of

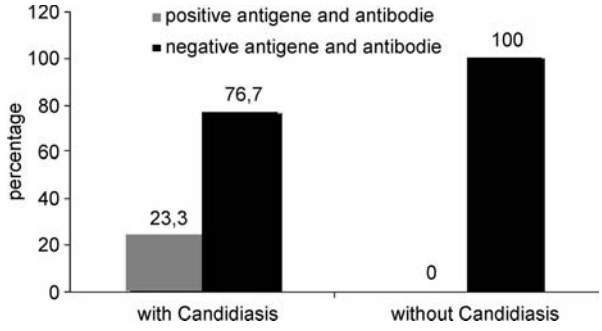

Fig. 5. Relation of positive results of antigens and antibodies in patients with and without clinical signs of Disseminated candidiasis.

patients who were positive to antigens and antibodies was higher ($p<0.01$) in the group of patients with positive hemoculture (3/7; 42.9%), in comparison to the group of patients who had negative hemoculture (4/53; 7.5%) (Figure 6).

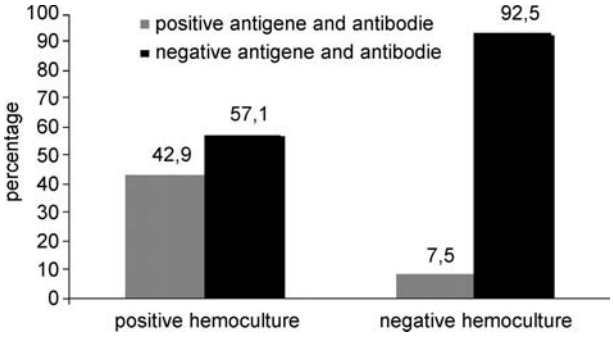

Fig. 6. Relation of positive results of antigens and antibodies and hemoculture.

Correlation of results of antigens and antibodies with hemoculture

The results of hemoculture and the results of mannan antigens and antibodies in patients with clinical signs of disseminated candidiasis were correlated. It was proved that there was statistically significant correlation between the results of hemoculture and mannan antigen ($\rho=0.36$; $p<0.05$), while the correlation of results be-

tween the hemoculture and antibodies was unsignificant ($\rho = 0.25$; $p = 0.19$). ROC (Receiver Operating Characteristics) analysis of diagnostic values of mannan antigen for prediction of positivity of hemoculture to *Candida* species was made (although this is a bad standard because *Candida* spp. is positive in 50% of cases). ROC space was 0.75 (%95CI = 0.65–0.84; $p<0.05$) and was statistically significant (Figure 7). The best diagnostic test performance on classifying positive hemoculture had predictive value 0,45, which had sensitivity of 57% and specifity of 98%, and PPV of 67% and NPV of 96%.

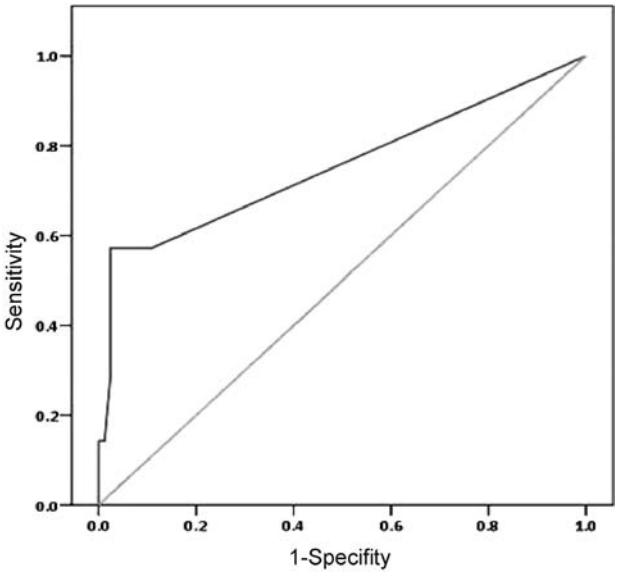

Fig. 7. ROC analysis of diagnostic values of mannan antigen for prediction of positivity of hemoculture to Candida species.

Discussion

Disseminated (systematic, invasive, hematogenic) candidiasis involves blood stream infections and inner organ infections, which are caused by penetration of *Candida* species yeasts into bloodstream. Frequency of these infections increases with the number of immunocompromissed patients, but it is also connected to invasive operations in departments of intense care. The incidence of candidemia is higher in Barcelona than in the USA (4.3 cases *per* 100 000 of the population) and it was 6 to 10 cases *per* 100 000 of the population[10]. The incidence of candidemia in North European countries is from 1.7 to 4.9 of cases *per* 100 000 of the population[11]. In comparison to results from USA the higher incidence rate is result of demographic factors and applying of different medical procedures. According to literature data in our country there were not any similar researches until today, which could give the exact estimate of incidence of candidiasis and its exposure. The percentage of the whole number of patients with candidemia in comparison to the number of patients with septicemia was 5% in the USA, and in European Countries it was 2–3%[12]. In Croatia, in 23.3% of patients with septicemia fungus etiology was confirmed[13]. In our reserach 11% of patients had candidemia 21% of patients had positive mannan antigen result which is, according to many authors, considered as indication of candidemia. Higher percentage of proved candidemia is explained by a good analyzed group. Fungus Infection Group at The European Organisation for Research and Treatment of Cancer (EORTC), has shown the difference in distribution of *Candida* species in patients with hematological malignities and in patients with solid tumors in their research. From 294 of patients 36% had oncological type of malignity and 64% had hematological type of malignity[14]. This research has shown that from 60 patients 61% of them had hematological type of malignity and 39% of patients had oncological type of malignity which matches with the literature data. Clinical signs and biochemical parameters showing the most predictive value in disseminated candidiasis diagnose are fever (body temperature higher than 38 °C) which do not react to antibiotic therapy and which has values of neutrophil leucocytes lower than 0.5×10^9/L. Fever lasting for 3 and more days and without any reaction to antibiotic therapy and values of neutropenia lower than 0.5×10^9/L is basic criteria for disseminated candidiasis diagnose[15]. In our research increased body temperature (>38 °C) lasting for 3 and more days and without any reaction to antibiotic therapy was one of the including signs for clinical diagnosis of disseminated candidiasis. Average body temperature in the group of patients with clinical signs of disseminated candidiasis was 38.8±0.40 °C. Average body temperature in the group of patients without clinical signs of disseminated candidiasis was 37.6±0.80 °C. It was approximately higher for 1.19 degrees (%95CI = 0.9–1.5), which was statistically significant difference ($p<0.001$). In both groups 2/60 of patients had 4 antibiotics in their therapy, 13/60 of patients had triple AB therapy, 15/60 of patients had double therapy nad 11/60 of patients had at least one antibiotic in their therapy. 19 of patients did not have any antibiotics in their therapy. Prolonged neutropenia (which takes longer than 10 days and with values $<0.5 \times 10^9$/L) is one of the most important factors which classifies patients with hematological malignities into the most risking group for getting disseminated candidiasis. In the research[15], 79% of patients had neutropenia ($<0.5 \times 10^9$/L) approximately for 20 days before the signs of disseminated infection, while in 11% of patients disseminated candidiasis was proved. In our research all patients with clinical signs of disseminated candidiasis had neutropenia which lasted longer than 10 days with values $<1.5 \times 10^9$/L. 43.3% of patients had neutrophil values $<0.5 \times 10^9$/L and from these in 15.3% of patients disseminated candidiasis was proved, which matches with the research's results. According to EORTC instructions from 1989, the usage of empiric antifungal therapy in neutropenic patients with fever lasting more than 5 days after applied antibiotic therapy, decreases the possibility of expansion of invasive fungal infections. In the research[13] have proved that 35% of patients had antifungal therapy before confirmed fungal infection. In our research, in the group of patients who did not have antifungal therapy 11.9% of patients had positive hemoculture. In the group of patients who had antifungal therapy 11.1% of patients

had positive hemoculture, which was statistically insignificant. The cause of this result lies in the fact that most authors, according to the literature data, recomend for antifungal prophylaxis amfotericin B and nystatin while our patients had flukonazol and nystatin prophylaxis. It was hard to do statistical analysis due to the low number of patients positive to hemoculture in the whole group. Statistically, more patients with positive antigen was found among the patients who did not have antifungal therapy (66.7%) in comparison to patients who had antifungal therapy (6.7%). There was not any significant difference in the presence of antibodies among the examined groups. Our results show positive effect of antifungal prophylaxis from preventing disseminated candidiasis. Due to the lack of specific signs and symptoms, disseminated candidiasis represents diagnostic and therapeutic problem[16]. Although hemoculture represents 'the golden standard' for disseminated candidiasis diagnose it also shows low sensitivity which goes around 50% and it usually takes several days to show detectable growth of *Candida*[17]. In the group of patients with clinical signs of disseminated candidiasis, 20% of patients had positive hemoculture. In the group of patients without clinical signs of disseminated candidiasis 3.3% of patients had positive hemoculture. The reason for low sensitivity is related to insufficient number of blood samples *per* one patient. Lately quick diagnostic tests, which at the beginning did not show high sensitivity and specificity, have developed. Diagnostic methods for detecting antibodies should differentiate candidal colonization of mucosa membranes or superficial tissue infections from candidemia, which requires antifungal therapy[18]. Specificity of antibodies detection tests can be low, due to the high titra in patients with colonized candida. The test can be with low sensitivity because the response of antibodies in immunocompromissed patients can be postponed, decreased or it even can disappear[19]. In order to improve sensitivity and specificity in detection of antibodies standard technique ELISA (Platelia *Candida* Ab) is used. Analyzing one serum sample *per* patient decreases the possibility to detect disseminated candidiasis, while analyzing more serum samples enables monitoring movement of titra antigen[20]. Sensitivity of anti-mannan and antibodies test was 47% and specificity was 100% with sample values cut-off >10 AU/mL. The level of antibodies from 15 positive serum samples was from 10.6 to 35 ($\overline{X}\pm SD = 19.81\pm 8.05$) AU/mL[21]. In our research we took one serum sample *per* patient and average value of titra antibodies in patients with positive hemoculture to *Candida* was 20.7 ± 16.1. The average value of titra antibodies in patients with negative hemoculture was 13.7 ± 15.5, which was not statistically significant ($p=0.94$). In our research average values of antibodies were higher ($p<0.05$) in the group of patients with clinical signs of disseminated candidiasis (15.85 ± 16.24), in comparison to the group of patients without clinical signs of disseminated candidiasis (13.24 ± 15.0). The difference was not large so we can conclude that values of antibodies can be increased in patients without disseminated candidiasis since the positive result can be colonization indicator. Mannan is the most important manoprotein of cell wall in *Candida* species. ELISA test enables disseminated candidiasis diagnose by detection of α-oligo mannose from serum which is released from cell walls during *Candida* infection. This test has good specificity but has to be repeated frequently because of the rapid clearance of mannan antigen from circulation[8]. Test sensitivity depends on the number of samples taken from each patient. In this research, sensitivity of mannanemia decreases from 40 to 11% if only one sample is examined per patient. The quantity of mannan antigen in serum of patients with positive hemoculture was from 0.5 to 3.19 ng/ml, ($\overline{X}\pm SD = 1.29\pm 0.88$), sensitivity of the test was 41% and the specificity of the test was 100%[21]. The research we took showed that the average value of titra antigen (0.86 ± 1.05) was statistically higher ($p<0.001$) in patients with positive hemoculture, in comparison to patients with negative hemoculture (0.13 ± 0.45). The approximate difference was 0.73 (%95CI = 0.30–1.17). Sensitivity of the test to mannan antigen was 57% and specificity was 98%. This values were simmilar to the values of Alam et al., since the same serological tests were used. After performing correlation test between hemoculture and mannan antigen and antibodies results in patients with clinical signs of disseminated candidiasis, statistically significant correlation was found ($\rho=0.36$; $p<0.05$), while the correaltion between hemoculture and antibodies was insignificant ($\rho=0.25$; $p=0.19$). According to our results and literature data detection of mannan antigen is more sensitive and faster method for disseminated candidiasis diagnose in comparison to hemoculture. New strategies for disseminated candidiasis diagnose combine determining values of manan antigen and antibodies in the serum applying ELISA technique (Platelia *Candida* Ag and Platelia *Candida* Ab). Retrospective studies have shown that sensitivity of these tests is 80 to 100%[9–19,22]. Combined use of both tests increases sensitivity from 80% to 95,4 and specificity from 52.6 to 93%[23]. In our research the number of patients who were positive to antigen and antibodies was higher ($p<0.01$) in the group of patients with clinical signs of disseminated candidiasis (23.3%), in comparison to the group of patients without clinical signs of disseminated candidiasis (0%). The number of patients who were positive to antigen and antibodies was higher ($p<0.01$) in the group of patients with positive hemoculture (42.9%),in comparison to the group of patients with negative hemoculture (7.5%). Mannan antigen detection serological tests have high negative predictive value (NPV) from 97%[8]. These high values, according to authors, are good indicators of Candidal infections and they are very useful in indetification of patients who need antifungal therapy. During our research ROC analysis for diagnostic values of mannan antigen in prediction to positive hemoculture to *Candida* species was performed and NPV was 96%, while PPV(positive predictive value) was 67%. Due to the low sensitivity of hemoculture and the time needed for isolation of *Candida* spp., introducing serological tests into regular procedures would speed disseminated candidiasis diagnose.

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ZNAČAJ PRIMJENE SEROLOŠKIH TESTOVA U DIJAGNOSTICI DISEMINIRANE KANDIDIJAZE

SAŽETAK

Diseminirana kandidijaza je definirana kao gljivična multiorganska infekcija uzrokovana kvasnicom iz roda *Candida* sa ili bez kandidemije. Učestalost diseminirane kandidijaze raste sa porastom broja imunokompromitiranih bolesnika. Posljednjih nekoliko desetljeća učestalost diseminirane kandidijaze je u porastu kao i stopa mortaliteta. Cilj ovog istraživanja je pokazati značaj primjene seroloških testova u dijagnostici diseminirane kandidijaze. Radi se o prospektivnoj studiji u koju je uključeno 60 pacijenata oboljelih od malignih oboljenja sa i bez kliničkih znakova diseminirane kandidijaze i 30 zdravih osoba koji čine kontrolnu grupu. Svim pacijentima smo pored kemokulture određivali prisustvo manan antigena i odgovarajućih antitijela C. species primjenom komercijalnog ELISA testa. U ovom istraživanju obrađeni su svi, prema dostupnoj literaturi, relevantni faktori za nastanak diseminirane kandidijaze. Ovo istraživanje je pokazalo da je u grupi pacijenata sa kliničkim znacima diseminirane kandidijaze značajno više ispitanika sa pozitivnom kemokulturom na Candidu species, nego u grupi pacijenata bez kliničkih znakova kandidijaze. Broj ispitanika koji su bili pozitivni i na antigen i antitijelo je bio značajno veći ($p<0,01$) u grupi ispitanika sa kliničkim znacima diseminirane kandidijaze (7/30; 23,3%), nego u grupi ispitanika bez istih (0/30; 0%). Prosječna vrijednost titra antigena je bila statistički značajno viša ($p<0,001$) u ispitanika sa pozitivnom, nego u onih sa negativnom kemokulturom. U grupi ispitanika sa kliničkim znacima diseminirane kandidijaze nađeno je ukupno 6/30 (20%) ispitanika sa pozitivnom kemokulturom na Candidu, dok je u grupi ispitanika bez kliničkih znakova nađeno 1/30 (3,3%), a što je bilo značajno više ($p<0,05$). Korelacijom nalaza kemokulture sa nalazom manan antigena i antijela u ispitanika sa diseminiranom kandidijazom nađena je statistički značajna korelacija, dok korelacija između nalaza kemokulture i antijela nije značajna. Zbog niske osjetljivosti kemokulture i potrebnog vremena za izolaciju *Candidae spp.*, uvođenje seroloških testova u redovnu proceduru znatno bi ubrzalo postavljanje dijagnoze diseminirane kandidijaze.