Novel Antifungal Drugs Against Fungal Pathogens: Do They Provide Promising Results for Treatment?

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Abstract The febrile neutropenia episodes of hematological patients and their outcomes were evaluated with respect to fungal pathogens and antifungal therapy in this retrospective study. All patients, who were older than 14 years of age and developed at least one neutropenic episode after chemotherapy due to hematological cancer from November 2010 to November 2012, were included into the study. We retrospectively collected demographic, treatment, and survival data of 126 patients with neutropenia and their 282 febrile episodes. The mean Multinational Association for Supportive Care in Cancer score was 17.18 ± 8.27 . Systemic antifungal drugs were initiated in 22 patients with 30 culture-proven invasive fungal infections (IFIs), 25 attacks of 19 patients with probable invasive pulmonary aspergillosis (IPA), 42 attacks of 38 patients with possible IPA, and 31 attacks of 30 patients with suspected IFI. Voriconazole (VOR), caspofungin and liposomal amphotericin B were used to treat 72 episodes of 65 patients, 45 episodes of 37 patients and 34 episodes of 32 patients as a first-line therapy, respectively. Unfavorable conditions of our hematology ward are thought to increase the number of cases with invasive pulmonary aspergillosis and VOR use. It should be taken into consideration that

increased systemic and per oral VOR usage predisposes patients to colonization and infection with azole-resistant fungal strains. Catheters should be removed in cases where patients' conditions are convenient to remove it. Acute myeloblastic leukemia cases that are more likely to develop invasive fungal infections should be monitored closely for early diagnosis and timely initiation of antifungal drugs which directly correlates with survival rates.

Keywords Hematological patients · Febrile neutropenia · Antifungal treatment · Invasive fungal infection · Fungemia

Introduction

Febrile neutropenia (FN) develops a mortal complication due to cancer chemotherapy [1]. Invasive fungal infections (IFIs) have been reported to have poor outcome in patients with prolonged neutropenia [2]. Early diagnosis of fungal infections and initiation of antifungal treatment are crucial for survival [3]. Diagnosis of the IFIs has been a big challenge in spite of new tests and modalities including computed tomography (CT) scanning and *Aspergillus* galactomannan antigen test that is recommended for pre-emptive antifungal therapy and reported to have a sensitivity rate of 58–65 % and a specificity rate of 65–95 % [1]. Mortality has declined between 5 % in patients with solid tumors and 11 % in those with hematological malignancies after new tests and modalities [4]. Profound neutropenia due to hematologic malignancy or allogeneic hematopoietic stem cell transplantation predisposes to invasive aspergillosis [4]. Empirical antifungal therapy is recommended to patients who have persistent neutropenic fever in spite of antibacterial therapy and have progressive

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pulmonary infiltrates associated with invasive aspergillosis by serial CT scanning to achieve a better outcome [4]. However, IFI was demonstrated in 4 % of patients who comprised only 22–34 % of the neutropenic patients who had cancer and received an antifungal drug according to established criteria [5].

Here, we retrospectively evaluated the FN episodes of hematological patients and outcomes with respect to fungal pathogens and antifungal therapy.

Materials and Methods

Patients, who were older than 14 years of age and developed at least one neutropenic episode after chemotherapy due to hematological cancer from November 2010 to November 2012 in the hematology ward of Ministry of Health Okmeydanı Training and Research Hospital in İstanbul, were included into the study. This study was approved by the local ethics committee. Patients, who were treated for other hematological diseases (anemia, idiopathic or immune thrombocytopenic purpura, etc.) and had not been screened with rectal swab culture for VRE colonization whilst residing in the hematology ward, were excluded. The hematology department is equipped with 23 beds. Patient rooms were designed as single, double and four-person without HEPA filters. FN was defined as an oral temperature $>38.3^{\circ}\text{C}$ or two consecutive readings of $>38.0^{\circ}\text{C}$ for 2 h and an absolute neutrophil count $<0.5 \times 10^9/\text{L}$, or expected count to fall below $0.5 \times 10^9/\text{L}$ [1]. Collected data included patient demographics and diagnosis, the episode data, clinical presentation and laboratory findings, clinical therapy, microbiological data and outcome. The FN treatment protocol was based on “Clinical Practice Guidelines for the Use of Antimicrobial Agents in Neutropenic Patients with Cancer” published in 2002 and the 2010 update by the Infectious Diseases Society of America (IDSA) [1, 6]. Empirical antifungal treatment was considered for patients with persistent or recurrent fever after 4–7 days of antibiotics and whose overall duration of neutropenia was expected to be more than 7 days. If CT showed changes associated with fungal infection, liposomal amphotericin B (L-AmB) or voriconazole (VOR) were initiated. VOR was continued as oral therapy after infection responded to systemic therapy and patient was able to receive oral therapy. Cavitation, air-crescent sign and halo sign were classified as the major changes for invasive pulmonary aspergillosis (IPA). Nodules and new infiltrates, including consolidation and effusions, were classified as minor changes [7]. Possible IPA was defined as clinical and radiologic findings highly suggestive of infection without established histopathologic and/or microbiologic evidence of infection. Probable IPA

was defined as a positive culture for *Aspergillus* species from a respiratory specimen or two consecutive *Aspergillus* galactomannan antigen tests with an index of ≥ 0.5 with clinical and radiologic findings suggestive of IPA. Proven IPA was defined as histopathologic evidence of tissue invasion and damage by *Aspergillus* species with clinical and radiologic findings [8]. First-line therapy was defined as given antifungal treatment initially. Salvage therapy was defined as an antifungal therapy which was given after antifungal treatment which did not achieve clinical and microbiological responses. POS was used for primary antifungal prophylaxis as given 200 mg per oral three times in a day with fat meal and acidic fruit juice during a period a time that neutrophil count decreased to below $1 \times 10^9/\text{L}$ subsequent to chemotherapy until recovered to $1 \times 10^9/\text{L}$. A scoring system for risk of complications among febrile neutropenic patients was based on Multi-national Association for Supportive Care in Cancer (MASCC) score [1]. Secondary antifungal prophylaxis was given to patients who were treated with IPA diagnosed clinically or microbiologically developed subsequent to previous chemotherapy as VOR 200 mg twice in a day per oral or POS 200 mg three times in a day during a period a time that neutrophil count decreased to below $1 \times 10^9/\text{L}$ subsequent to chemotherapy until recovered to $1 \times 10^9/\text{L}$. If patient could not receive oral therapy, secondary antifungal prophylaxis was given intravenously. Antibiotic prophylaxis was not given to any patients. Blood samples drawn from a vein or catheter were inoculated into Bact-Alert 3D bottles (bioMérieux Diagnostics, Lyon, France). In addition, samples of urine, sputum, wound, conjunctive, abscess, blood and catheter were inoculated onto 5 % sheep-blood agar (Salubris Inc., İstanbul, Turkey), or chocolate agar (Salubris Inc., İstanbul, Turkey) and MacConkey agar (Salubris Inc., İstanbul, Turkey). Isolated yeasts from blood cultures were identified by morphological examination on Sabouraud dextrose agar plates (Unipath Ltd., Basingstoke, England), examination of germ-tube formation and API ID 32C (bioMérieux Diagnostics, Lyon, France) for discrimination at the species level. The ATB Fungus 2 microdilution kit (bioMérieux Diagnostics, Lyon, France) was used for susceptibility testing according to the CLSI (formerly NCCLS) broth microdilution M27-A2 protocol. *Aspergillus* galactomannan antigen test was performed using Platelia commercial enzyme immunoassay kit (Platelia *Aspergillus* EIA; Bio-Rad Laboratories, Marnes-la-Coquette, France). Blood samples of patients were analyzed twice weekly and results were computed as an index in which values of ≥ 0.5 were considered as positive relative to an optical density of the control sample measured with a semiautomatic analyzer (Behring ELISA processor III; Dade Behring, Marburg, Germany) [9]. Possible causes of false-positive and negative results were

eliminated. The test was considered significant if the patient had two consecutive *Aspergillus* galactomannan antigen tests with an index of ≥ 0.5 under persistent or recurrent fever after 4–7 days of antibiotics with or without microbiological or radiological findings associated with fungal infections as mentioned earlier and whose overall duration of neutropenia was expected to be more than 7 days [1]. Catheters were removed if the patient did not respond to antifungal therapy with deteriorating clinical, radiological and laboratory findings. Catheters were not removed in patients who needed to central line to receive treatment intravenously, due to trouble with intravenous route. Nephrotoxicity was defined as an increase in serum creatinine value greater than two times the baseline value or with a peak >2.0 mg/dL [10]. Hypokalemia was defined as a serum potassium level <3.5 mmol/L (<3.5 mEq/L). Moderate hypokalemia is defined as serum potassium levels of 2.5–3 mmol/L (2.5–3 mEq/L) and severe hypokalemia defined as a serum potassium level <2.5 mmol/L (<2.5 mEq/L) [11]. Blood concentration of VOR could not be monitored due to unavailability of analysis in Turkey.

Response to treatment was defined as recovery of laboratory, radiological findings, vital signs and clinical symptoms associated with infection (improvement in arterial blood-gas values, radiological improvement, negative urine culture for urinary system infection and recovery of signs and symptoms related to other infections). In-hospital mortality during the neutropenic phase within 30-day and the clinical outcomes of febrile neutropenic episodes associated with IFI were the primary consequences that were investigated in this study.

Statistical Analysis

Continuous variables were described as mean $\pm$ SD and range. Percentile values were described without decimals. Overall 30-day crude mortality associated with FN was defined as death within 30 days after development of neutropenia. Crude 30-day mortality rates were calculated as the proportion of patients who died within 30 days after development of neutropenia to all patients who were included into the study. Mortality rates related to IFI were calculated as the proportion of patients who died of IFI that developed in the neutropenic phase and sustained after recovery of neutropenia within 30 days to IFI attacks of all patients who were included into the study.

Results

In this retrospective, observational study, we analyzed 282 febrile episodes of 126 consecutive patients with neutropenia comprising 87 cases in the first year and 78 cases in

the second year. Mean age was 51.7 ± 14.4 years (range 17–82 years) and 66 cases were male. Mean MASCC score and duration of neutropenia were 17.18 ± 8.27 and 29.38 ± 6.95 in the patients with hematological malignancies, respectively (Table 1). Systemic antifungal drugs were initiated in 22 patients with 30 culture-proven IFIs (Table 2), to 19 patients with probable IPA in 25 attacks, to 38 patients with possible IPA in 42 attacks, and to 30 patients in 31 attacks with suspected IFI. VOR, caspofungin (CAS) and L-AmB were used to treat in 72 episodes of 65 patients, 45 episodes of 37 patients and 34 episodes of 32 patients as a first-line therapy, respectively (Table 2). Of 30 culture-proven IFIs in 22 cases, 21 attacks of 14 cases developed in the first year and 9 attacks of 8 cases developed in the second year (Table 3). Five patients were treated with CAS due to hepatosplenic candidiasis depending on clinical and radiological findings. Central venous catheter could be removed from 10 of 20 patients whose blood cultures yielded yeast and four of those (40 %) died (Table 3). Of 13 patients who received primary antifungal prophylaxis, six (43 %) patients did not need systemic antifungal treatment under induction or consolidation chemotherapy and one patient with colostomy could not tolerate it due to diarrhea. Of 23 patients who received secondary antifungal prophylaxis with per oral either VOR or POS in 45 episodes, systemic antifungal therapy was not needed in 22 of 38 episodes and five of eight episodes, respectively. Secondary antifungal prophylaxis with CAS was found effective in six episodes of three patients who had at least two persistent candidemia attacks (Table 2). The treatment with VOR was discontinued among 65 patients who received VOR; in 10 patients (15 %) due to hallucination under per oral or intravenous treatment, in three patients (4 %) due to elevated liver enzymes and in two patients (3 %) due to blurred and colored visual change. L-AmB was discontinued in six patients (18 %) due to life-threatening severe hypokalemia and nephrotoxicity. All patients who received L-AmB received potassium supplement. Posaconazole (POS) was discontinued due to diarrhea ($n = 2$, 8 %), elevated liver enzymes ($n = 1$, 4 %) among 25 patients who received POS. There was no side-effect related to CAS except for one patient who developed an allergic reaction after initiation. There was no proven infection associated with *Zygomycetes* or *Fusarium* species. Previous azole exposure was identified in 13 of 18 cases with bloodstream yeast infections. In total, eight patients with acute myeloid leukemia and two patients with acute lymphocytic leukemia who had findings in context with IFIs were treated with a combination of antifungal drugs as salvage therapy due to persistent fever and negative culture, progression in thorax CT findings and deterioration. Five of those (50 %) responded to combination therapy.

Table 1 Characteristics and distribution of hematologic malignancies in patients with febrile neutropenia (n = 126)

Patients	
Gender (male/female)	66/60
Mean age	51.7 ± 14.4
Mean MASCC score	17.18 ± 8.27
Mean duration of neutropenia	29.38 ± 6.95
n (%)	
Hematologic malignancies	
Acute myeloblastic leukemia	73 (58)
Acute lymphocytic leukemia	22 (17)
Non-Hodgkin lymphoma	7 (5)
Chronic lymphocytic leukemia	5 (4)
Multiple myeloma	5 (4)
Hairy cell leukemia	4 (3)
Aplastic anemia	3 (2)
Chronic myeloid leukemia	2 (2)
Plasma cell leukemia	2 (2)
Mantle-cell lymphoma	2 (2)
Chronic lymphocytic leukemia with Burkitt's lymphoma	1 (1)
Total	126 (100)

VOR and L-AmB combination were used in three episodes of three patients. Two of three patients responded to treatment, whereas others could not tolerate the treatment due to hallucination which is known a side effect of VOR. VOR and CAS combination were used in four episodes of four patients empirically and none of them responded to treatment. L-AmB and CAS combination were used in four episodes of three patients empirically, two of whom responded to treatment in three episodes. Per oral VOR and POS therapies achieved clinical responses as maintenance antifungal therapy in 16 and 18 patients at least 4 weeks after systemic antifungal therapy without toxicity and discontinuation, respectively. Of five patients who had pulmonary findings in thorax CT and received empirical CAS therapy, four patients achieved clinical and radiological responses after switching to VOR and so did one patient after switching to L-AmB. Overall 30-day crude mortality rates were 26 % (23/87) among patients who were diagnosed with AML (n = 16), acute lymphocytic leukemia (ALL, n = 5), multiple myeloma (n = 1), chronic myeloid leukemia (n = 1) in the first year and 21 % (17/78) among patients who were diagnosed with acute myeloblastic leukemia (AML, n = 16), acute lymphocytic leukemia (ALL, n = 4), non-Hodgkin lymphoma (n = 1) in the second year. The number of patients who died of infections was 17 (19 %) in the first year, 11 (10 %) in the second year. Mortality rates related to IFI were 5 % (n = 5) in the first

year, 3 % (n = 3) in the second year and 6.3 % (n = 8) in overall, respectively (Table 2). Mortality rates were 11 % (n = 1/9) in the first year, and 22.2 % (n = 2/9) in the second year for candidemia. Bacterial infections, including pneumonia, urinary tract infections, central-line blood stream infections, intraabdominal infections, and both vancomycin sensitive *Enterococcus faecalis* and vancomycin resistant *E. faecalis* that induced severe sepsis were also recorded in patients who died.

Discussion

IFIs cause to mortality in patients with hematologic malignancies due to *Aspergillus* and *Candida* species [12]. Mean MASCC score and duration of neutropenia of our patients and unfavorable conditions of our hematology ward, such as absence of single rooms with HEPA filter and private toilet, are thought to increase the number of cases with invasive pulmonary aspergillosis and VOR usage than other antifungal drugs in more than half of all febrile neutropenic episodes [151 of 282 (53 %) episodes] in context with possible or probable IPA that predominated among IFIs [13]. However, empirical antifungal therapy was reported to be initiated to 67 % of patients followed-up at ward with low environmental contamination in microbiological surveillance [14]. VOR was mostly initiated antifungal drug in context with the number of patients diagnosed with probable and possible IPA compared to L-AmB. The presence of oral formulation, lower price compared to L-AmB and guideline recommendations lead to initiation of VOR. CAS did not achieve expected clinical response in our patients with possible or probable invasive pulmonary infections who responded to salvage therapy with VOR (n = 4) to L-AmB (n = 1). CAS has been described as second-line agent for treatment of invasive aspergillosis in patients who cannot tolerate VOR or need enhanced antifungal activity combined with VOR. CAS was found to be effective as empirical or pre-emptive therapy and secondary prophylaxis in our patients who had one of those including candidemia, persistent candidemia attacks, hepatosplenic candidiasis [15]. Antifungal combination therapy is still controversial and has not been demonstrated evidently. Toxicity and cost are major concerns regarding antifungal combination therapies. IDSA guidelines recommended antifungal combination therapy as second-line (salvage) [16]. Antifungal combination therapies including VOR combined with L-AmB and CAS combined with L-AmB achieved clinical response as salvage therapy in our study. The cases did not respond to VOR plus L-AmB combination therapy as reported by previous studies [17, 18]. In COMBISTRAT trial, the combination of standard dose L-AmB (3 mg/kg/day) plus

Table 2 Antifungal therapies and prophylaxis given to patients and mortality rates related with IFIs

	Number of cases, attacks
Distribution of cases given antifungal therapy	
Culture proven	22, 30
Probable IPA	19, 25
Possible IPA	38, 42
Suspected IFI	30, 31
Hepatosplenic candidiasis	5, 5
First-line antifungal therapy	
With VOR	65, 72
With CAS	37, 45
With L-AmB	32, 34
Salvage therapy with combined antifungal therapy	8, 8
VOR plus L-AmB	3, 3
VOR and CAS	4, 4
L-AmB and CAS	3, 4
Primary POS prophylaxis	13, 13
Secondary prophylaxis	23, 45
With VOR	17, 35
With POS	6, 10
With CAS	3, 6
	n (%)
Discontinuation of VOR (n = 65)	
Hallucination	10, 15
Elevated liver enzymes	3, 4
Blurred and colored visual change	2, 3
Total	15, 23
Discontinuation of L-AmB	6, 18
Discontinuation of CAS (n = 37)	1, 2
Discontinuation of POS (n = 25)	
Diarrhea	2, 8
Elevated liver enzymes	1, 4
Total	3, 12
Overall 30-day crude mortality rates	
First year	23, 26
Second year	17, 21
Mortality rates due to infection	
First year	17, 19
Second year	11, 10
Mortality rates due to IFIs	
First year	5, 5
Second year	3, 3
Overall in 2 years	8, 6
Due to candidemia in the first year	1, 11
Due to candidemia in the second year	2, 22
Overall mortality for 2 years	3, 16

VOR voriconazole, L-AmB liposomal amphotericin B, CAS caspofungin, FLC fluconazole, ITR itraconazole, AmB amphotericin B, IFI invasive fungal infection, IPA invasive pulmonary aspergillosis

CAS achieved higher response rates 67 % that did high dose L-AmB (10 mg/kg/day) with 27 % in 30 patients with hematologic malignancies and proven or probable invasive

aspergillosis [19]. Survival rates at 12 weeks were similar, 100 % for the combination and 80 % for monotherapy. But toxicity developed more in those cases with combination

Table 3 Isolated fungal pathogens and response to antifungal treatment in patients with febrile neutropenia

Patient	Age	Gender	Hematologic malignancy	Sample	Isolated fungal pathogen	Antifungal resistance	Previous azole exposure	Empirical antifungal treatment before identification	Treatment modification	Outcome
1	32	Male	AML	Blood	<i>Candida parapsilosis</i>	VOR, FLC	+	CAS	–	Survived
				Blood	<i>Candida parapsilosis</i>	VOR, FLC	+	CAS	–	Survived
				Blood	<i>Candida glabrata</i>	FLC	+	L-AmB	Central line removed	Survived
2	51	Male	AML	Blood	<i>Candida parapsilosis</i>	VOR, FLC	+	CAS	Central line removed	Survived
				Blood, wound	<i>Candida parapsilosis</i>	VOR, FLC	+	VOR	From VOR to AM-B	Died
3	32	Male	AML	Blood	<i>Candida parapsilosis</i>	VOR, FLC	+	CAS	–	Survived
				Blood	<i>Candida parapsilosis</i>	VOR, FLC	+	VOR	L-AmB	Survived
4	52	Female	ALL	Blood	<i>Geotrichum capitatum</i>	FLC, ITR	–	CAS	Central line removed	Survived
				Blood	<i>Geotrichum capitatum</i>	FLC, ITR	+	VOR	–	Died
5	59	Female	AML	Urine, sputum	<i>Candida kefyr</i>	FLC	+	L-AmB	–	Survived
6	60	Female	ALL	Blood	<i>Candida albicans</i>	VOR, ITR, FLC	–	CAS	–	Survived
7	21	Female	Aplastic anemia	Blood	<i>Candida albicans</i>		–	CAS	–	Survived
8	33	Male	AML	Blood	<i>Geotrichum capitatum</i>	FLC	–	VOR	Central line removed	Survived
9	60	Female	AML	Sputum	<i>Candida kefyr</i>	FLC, ITR	+	VOR	–	Survived
				Sputum	<i>Candida kefyr</i>	FLC, ITR	+	VOR	–	Died
10	58	Female	CLL + Burkitt's lymphoma	Sputum	<i>Aspergillus fumigatus</i>		–	CAS	–	Survived
11	59	Female	AML	BAL	<i>Aspergillus fumigatus</i>		–	VOR	–	Survived
				Sputum	<i>Aspergillus fumigatus</i>		+	L-AmB	–	Survived
12	55	Female	AML	Blood	<i>Trichosporon asahii</i>	AmB	+	CAS	Central line removed	Died
13	63	Female	AML	Blood	<i>Trichosporon asahii</i>	AmB	+	L-AmB	From AM-B to VOR, central line removed	Died

Table 3 continued

Patient	Age	Gender	Hematologic malignancy	Sample	Isolated fungal pathogen	Antifungal resistance	Previous azole exposure	Empirical antifungal treatment before identification	Treatment modification	Outcome
14	45	Female	AML	Blood	<i>Trichosporon asahii</i>	AmB, ITR	–	CAS	Empirically switched to VOR	Died
15	50	Male	AML + lung cancer	Blood	<i>Candida parapsilosis</i>		+	VOR	From VOR to AM-B, central line removed	Survived
				Blood	<i>Candida albicans</i>	–	+	VOR	–	Survived
16	62	Female	AML	Blood, sputum	<i>Candida parapsilosis</i>		+	VOR	From VOR to CAS, central line removed	Died
17	61	Male	AML	Blood, Urine	<i>Candida albicans</i>		+			Died
18	73	Female	NHL	Blood, central line	<i>Candida albicans</i>		+	L-AmB	Central line removed	Survived
19	35	Male	AML	Blood	<i>Candida albicans</i>		+	CAS		Survived
20	44	Female	ALL	Blood	<i>Candida albicans</i>		+	L-AmB		Survived
21	63	Female	AML	Blood	<i>Candida krusei</i>		+	VOR	–	Survived
22	51	Female	AML	Blood	<i>Candida albicans</i>	–	+	VOR	–	Survived

AML acute myeloblastic leukemia, ALL acute lymphocytic leukemia, CLL chronic lymphocytic leukemia, NHL non-Hodgkin lymphoma, BAL bronchoalveolar lavage, VOR voriconazole, L-AmB liposomal amphotericin B, CAS caspofungin, FLC fluconazole, ITR itraconazole, AmB amphotericin B

therapy than those with monotherapy. VOR related hallucination developed in cases with VOR plus L-AmB combination therapy. In a study of Rojas et al. [20], treatment response rates and 12-week survival rates were similar in cases with three combination therapies including L-AmB plus CAS, L-AmB plus a triazole (VOR 16 and POS 4), and VOR plus candidin (20 CAS and 1 anidulafungin) for invasive mold infections. Survival rates were found similar in patients with primary antifungal combination therapy and patients with initial monotherapy (22 vs. 32 %) [21]. It could be given as salvage therapy to patients with invasive mold infections and persisting or progressive findings under monotherapy as in our study. As it is known that antifungal therapy is expensive, especially when administered systemically, oral POS and VOR could be regarded as cheaper options for maintenance therapy. In total, 34 patients recovered with maintenance oral antifungal therapy in our study. The blood concentration of those drugs could not be monitored in our study, but monitorization of drug concentration increases efficacy and decreases the risk for toxicity since blood concentrations of drugs could not reach to therapeutic levels in some patients [22, 23]. Main problem observed with orally administered drugs is the emergence of azole resistance in *Aspergillus* and *Candida* species and related infections among patients who received those drugs as mentioned earlier [24]. Either L-AmB or CAS should be evaluated for treatment if an IFI develops during or after treatment [25, 26]. VOR was reported to be inappropriate for 40 % of patients received as targeted, empirical, pre-emptive, prophylactic [23]. Patients with maintenance therapy with oral either VOR or POS should be followed up closely. Patients with proven IFI was reported to need antifungal therapy switching compared to patients with probable or possible IFI (33 vs. 25 and 7 %, respectively) [27].

Candida albicans (nine cases) predominated among fungal pathogens responsible for blood stream infections in our study as reported in other studies [28]. Two patients who died of *Candida parapsilosis* related blood stream infection yielded this species in sputum and wound cultures that revealed colonization. The previous azole exposure that was recorded for most of the cases in our study caused colonization of the mucosal membranes by *Candida* species preceded invasive diseases [25]. *Candida kefyr* was identified as resistant to fluconazole, and itraconazole in patient with history of VOR therapy in our study. Moreover, *Candida kefyr* remained to growth in the second sputum of the patient who died under VOR therapy (Table 3). *Candida* colonization can cause to candidemia in case mucosal barriers are impaired by chemotherapy preceding fatal outcome [29]. *Trichosporon asahii* (three cases) and *Geotrichum capitatum* (two cases) have emerged causing to death among our patients with blood

stream infections. *Trichosporon* was reported as the second most common cause of disseminated yeast infections which can also cause to break through under CAS treatment as in our study [30]. Although catheter removal is recommended after identifying the fungemia, catheter removal by 24 or 48 h after treatment initiation was reported to have no effect on overall treatment response, mortality and mycological eradication in 842 patients with candidemia [31–33]. CAS therapy is recommended by ESCMID Fungal Infection Study Group if catheter retention is needed due to clinical conditions of the patient [34]. The catheter was removed in cases where patients' conditions were convenient to remove it. Catheter removal should be taken into consideration, especially in cases with either *Trichosporon asahii* and *Geotrichum capitatum* owing to the fact that those cause to fatal outcomes as in our study.

Azole resistance has emerged among *C. parapsilosis* and *C. albicans* isolated from our patients. Especially previous azole exposure rates elucidate the resistance problem among our cases (Table 2). VOR was the most frequently used antifungal drug predisposing to colonisation of azole-resistant fungal pathogens in those patients. VOR resistance has been reported to emerge recently under VOR treatment from 0.3–10 % in different studies. Since frequent use of VOR was reported to be related to both probable and possible *Aspergillus* infections [35–37]. Either L-AmB or CAS is recommended for the treatment of fungemia caused by azole-resistant *Candida* species. Periodic shift of anti-fungal drugs, such as VOR with L-AmB for *Aspergillus* infections, CAS with L-AmB for empirical antifungal treatment, etc. could be evaluated, but prospective, randomised-controlled studies are needed regarding this subject.

IFI cases were nearly one-third of fatal infectious cases. Fatality rates of IFI among our patients (5 % in the first year, 3 % in the second year) were similar to those reported in previous studies. IFI cases decreases in the second year accompanying with the least number of fatal IFI cases. AML cases who consisted the majority of all our cases and fatal cases were vulnerable to IFI accompanying with environmental unprotective conditions. IFI cases were reported to be the principal cause of death (42/121, 35 %), followed by a relapse of malignancy (28 %) and other causes (37 %) [27]. In that study, patients whose antifungal therapy was not modified had better survival (47 vs. 31 % at 1 year, 29 vs. 9 % at 3 years) than patients whose antifungal therapy was modified. Only proven IFI was reported to increase mortality significantly (hazard ratio 1.67). Sex, diagnosis, treatment modality, underlying disease were found to be insignificant for survival as well [27]. Overall 1- and 3-year survival rates were reported to be significantly better in patients with probable or possible

IFI than patients with proven IFI (45, 46 vs. 32, and 25, 32 vs. 9 %, respectively) [27].

Conclusion

Unfavorable conditions of our hematology ward are thought to increase the number of cases with IPA and VOR usage. It should be taken into consideration that increased systemic and per oral VOR usage predisposes patients to colonization and infection with azole-resistant fungal strains. Catheters should be removed in cases where patients' conditions are convenient to remove it. AML cases that are more likely to develop IFI should be monitored closely for early diagnosis and timely initiation of antifungal drugs that correlate with better outcomes for survival.

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