Impact of Fluconazole versus Posaconazole Prophylaxis on the Incidence of Fungal Infections in Patients Receiving Induction Chemotherapy for Acute Myeloid Leukemia

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Background: Invasive fungal infections (IFIs) remain one of the worrying complications in patients with acute myeloid leukemia (AML) due to their incidence and high level of attributable mortality. In light of these risks, antifungal prophylaxis has always been debated. We conducted a single-center retrospective study of two prophylactic antifungal agents (fluconazole/posaconazole) in 91 consecutive patients receiving induction chemotherapy for AML between 2005 and 2009, in order to evaluate the impact on the incidence of IFI and on the mycological flora of the patients.

Methods: In total, 39 patients received prophylactic fluconazole versus 52 who received posaconazole. The baseline characteristics of the two groups were comparable.

Results: Overall, 17 patients developed an IFI, with no difference in frequency between the two groups. Utilization of empirical or pre-emptive therapy was similar irrespective of the type of prophylaxis used. Mycological examination of stools revealed an increase in non-*albicans Candida* colonization in the fluconazole group during hospitalization and the appearance of *Saccharomyces cerevisiae* colonization in patients receiving posaconazole.

Conclusion: The present study does not distinguish between fluconazole and posaconazole as a primary effective prevention against fungal infections. More prospective studies and meta-analyses are warranted.

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At a Glance Commentary

Scientific background of the subject

In 2009, the ECIL has recommended larger spectrum prophylaxis using posaconazole in high-risk patients with acute myeloid leukemia or myelodysplastic syndrome receiving intensive chemotherapy. With the authorization to market posaconazole, our old prophylactic regimen using fluconazole was replaced with one using posaconazole. Evaluations of posaconazole treatments are needed to further gather data on this matter.

What this study adds to the field

Our protocols to empirically treat invasive fungal infection were modified to use posaconazole. We evaluated whether these changes of practice had an impact in terms of patient management. Our results found no difference between posaconazole and fluconazole in contrast with other recent studies.

Key words: acute myeloid leukemia, antifungal prophylaxis, invasive fungal infections

Fungal infections are related to human communal agents or to environmental saprophytes and are rare in healthy subjects. Known contributing factors are reduced defense mechanisms of the host, the use of certain

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treatments (chemotherapy, corticosteroids, large-spectrum antibiotics, parenteral feeding, etc.), foreign objects (notably blood and urinary catheters), as well as the presence of fungi in the environment (*Aspergillus* and work).[1,2]

In patients with hematological malignancies, the incidence of invasive fungal infections (IFIs) has increased regularly during the last decade. The majority of these infections occur in patients with acute myeloid leukemia or myelodysplastic syndrome (MDS) or in those who have received allogenic hematopoietic stem cell transplants.[3,4] In patients with acute leukemia, the incidence of proven/probable IFI is estimated to be 24% (varying from 2 to 40% depending on the center).[5-7] The predominant species involved in Europe are *Candida* sp. and *Aspergillus* sp. However, in patients with hematological malignancies, *Aspergillus* species are the most frequent causal agents.[8-10] The mortality rate attributable to IFI is estimated to be between 6% and 60%, depending on the type of infectious agent. Due to their frequency and severity, IFIs increase the duration of hospitalization and, hence, the hospitalization costs. In light of the consequences in these high-risk patients, large-spectrum prophylaxis is often recommended.[11,12] In addition, it is actually admitted that the delay in instituting an adequate therapy is a negative factor in terms of mortality. Due to this, empiric therapy is frequently instituted in view of the slightest clinical suspicion.[13,14] The principal consequence of this practice is the difficulty in identifying the causal agent in more than 50% of fungal infections due to negative cultures[15,16] as well as the emergence of resistant fungi (non-*albicans Candida, Aspergillus flavus* or *terreus, Fusarium* sp., and *Zygomycetes* sp.).[17-22] Finally, pre-emptive therapy, targeting *Aspergillus*, would limit the over-treatment of the patients, thereby reducing the emergence of resistance, debilitating side effects, and costs. However, this approach is not yet the standard of care.[23-26]

In 2007, the European Conference on Infections in Leukemia patients (ECIL) proposed molecules such as fluconazole or itraconazole as preventive antifungals, for which the major drawbacks are the absence of efficacy for *Aspergillus* sp. and/or intolerance.[27,28] In 2009, the ECIL has recommended larger spectrum prophylaxis using posaconazole in high-risk patients with AML or MDS receiving intensive chemotherapy.

With the authorization to market posaconazole, our old prophylactic regimen using fluconazole was replaced with one using posaconazole. Our protocols to empirically treat IFI were also modified. We, therefore, wanted to evaluate whether these changes of practice had an impact in terms of patient management.

METHODS

Patients

The population included in this study consisted of patients aged 18 years and older, with a primary diagnosis of AML or MDS, who were hospitalized in our unit between 2005 and 2009 for induction chemotherapy to produce prolonged neutropenia (>10 days) and who received prophylactic fluconazole or posaconazole against IFI.

Study outline

This is a retrospective, pre-post comparative study involving 91 consecutive patients. During the first period of the study (2005-2007), all patients (n = 39) received prophylactic antifungal therapy consisting of fluconazole 400 mg p.o., with or without a non-absorbable gut decontamination, beginning on the first day of the induction chemotherapy. All patients received an induction treatment sequence based on anthracycline (idarubicine for the majority of patients) and aracytine intravenously continuously during 24 h. Dose and duration were fixed according to age (notably in the case of age up to 60 years). During the second period of the study (2007-2009), all patients (n = 52) received prophylactic antifungal therapy consisting of posaconazole 200 mg p.o. t.i.d., with or without a non-absorbable gut decontamination, beginning on the first day of the induction chemotherapy. The first febrile episode was treated with a broad-spectrum antibiotic as monotherapy after investigation of the causative agent. The persistence of fever after 72 h or the occurrence of a new febrile episode, non-documented, at more than 72 h after initiation of the antibiotic therapy resulted in the initiation of an empirical antifungal therapy consisting of caspofungin for the fluconazole group and liposomal amphotericin B (3 mg/kg/day) for the posaconazole group.

During the two study periods, all patients (except two) were lodged in individual rooms equipped with HEPA particle filters and positive pressure. All patients received vacuum-conditioned food. They underwent mycological follow-up during their stay, which consisted of direct examination and culture of buccal lavage and a daily sampling of stools. The search for antigens and serology for *Aspergillus* were performed twice weekly. The same examinations were conducted upon the readmission of each patient during follow-up treatment.

Suspicion of clinical or radiological pneumonia or a positive *Aspergillus* antigen test resulted in a computed tomography (CT) scan and a change in the prophylactic or empiric antifungal therapy to pre-emptive therapy with voriconazole (400 mg b.i.d. i.v. for 48 h followed by 200 mg

b.i.d. p.o.).[29,30] Depending on the pulmonary condition and the patient's clinical status, a broncho-alveolar lavage (BAL) was performed for bacteriological and mycological testing.

This retrospective non-interventional observational study obtained confidential approval from the Commission nationale de l'informatique et des libertés (CNIL). Local ethics board approval was obtained for the analysis of patient data. Helsinki convention criteria were respected for the purposes of the present study. Informed consent of patients was not required as data were anonymized.

Definitions

Severe neutropenia was defined as a polynuclear neutrophil count of < 0.5 g/l. The karyotype was classified into three categories as good, intermediate, or poor prognosis, according to the classification of Grimwade.[31] IFIs were defined as proven, probable, or possible in conformity with the European Organization for Research and Treatment of Cancer (EORTC) criteria.[32] The galactomannan test was designated positive at a value > 0.5.[33]

Cost analysis

The study took into account the hospital costs related to the prophylactic, empirical, and pre-emptive therapy for IFI. The costs used for the years before 2009 were actualized according to the annual index of hospital costs. Specifically for fluconazole, which became generic between 2007 and 2008, the price actualization was approached differently in order to respect the between-group comparison. The other associated costs were ignored under the hypothesis that the duration of hospitalization, nursing costs, and the cost of concomitant treatments should have been equivalent for the two groups. The unit costs (per day) for the antifungals for each year were taken from the hospital pharmacy for all the drugs used.[34]

Outcome criteria

The principal outcome criterion was the incidence of proven or probable fungal infections recorded at the end of the neutropenia induced by the chemotherapy. Secondary criteria included the number of possible IFIs, the duration of prophylactic and empirical therapy, the incidence of associated bacterial infections, the evolution of the mycological flora of the patients, as well as the effect of prophylaxis on colonization. The costs of the antifungal therapies were also evaluated.

Statistical analysis

The statistical analysis was conducted using SAS®, version 9.2 software. A univariate analysis was performed for all the variables which could have been affected by prophylactic treatment, and the comparisons were carried out using the Chi-squared or the Fisher's test for qualitative variables and the Wilcoxon–Mann–Whitney test for quantitative variables. All patients were followed up to the end date of the study (31 July 2010) or until death if it occurred prior to the end date of the study. Global survival was estimated using the Kaplan–Meier method, and the stratified curves for prophylaxis and the IFIs were analyzed using the log-rank test. Finally, a multivariate analysis was performed using the Cox model.

RESULTS

General characteristics of the population

The general characteristics of the population are given in Table 1. In total, 91 patients received prophylactic therapy. Thirty-nine (43%) received fluconazole and 52 (57%) received posaconazole. The mean age was 56 years (range 23–80 years). The principal demographic variables were similar for the two groups in terms of sex, medical history, type of pathology, presence of hyperleukocytosis at diagnosis (30% of patients), and neuro-meningeal invasion (4% of patients). The presence of neutropenia at admission was more frequent in the fluconazole group (44% vs 29%, p = NS). Hospitalization prior to admission to the isolation unit occurred in 25 patients (48%) in the posaconazole group versus 10 patients (26%) for the fluconazole group ($p = 0.03$). Two-thirds of the cases arrived from the emergency unit.

Overall, 89% of patients were in complete remission at the end of therapy, with 15% requiring remedial chemotherapy on Day 15. The mean duration of neutropenia was 28.7 days (range 8–81 days) for fluconazole patients and 32.2 days (range 12–182 days) for those receiving posaconazole (p = NS).

Infectious characteristics of the population

Infectious characteristics of the population are presented in Table 2. During hospitalization, each patient received between 0 and five different antibiotics, with a mean of 2.8 antibiotics per patient in the fluconazole group and 2.6 antibiotics per patient in the posaconazole group (p = NS). The average duration of prophylaxis was 14.4 days, which was comparable between the two groups. Thereafter, 80% patients received an empirical antifungal therapy consisting of caspofungin for 32 patients (82%) receiving fluconazole and liposomal amphotericin B for 39 posaconazole patients (75%). The primary cause for instituting empirical treatment was a new febrile episode. Only five fluconazole patients and nine posaconazole patients received just the prophylactic treatment. Concerning the pre-emptive therapy

for invasive pulmonary aspergillosis (IPA), 35% ($n = 32$) of patients received voriconazole.

Forty-two pulmonary CT scans were performed in 37 patients, and 29% were compatible with a diagnosis of IPA. The second CT scan performed for five patients permitted the readjustment of the diagnosis from non-specific pneumonia to probable IPA in two cases. Six fluconazole patients and seven posaconazole patients received a bronchial fibros-

Table 1: General characteristics of the population

	Fluconazole (*n*=39)	Posaconazole (*n*=52)	*p*
Age (years)	56.1 (29-80)	56.2 (23-75)	0.98
Sex			
Male	26 (66.7)	31 (59.6)	0.49
Female	13 (33.3)	21 (40.4)	
Medical history			
Diabetes	4 (10.3)	5 (9.6)	1.00
Cardiac insufficiency	2 (5.1)	1 (1.9)	0.57
Arrhythmia	0 (0.0)	3 (5.8)	0.26
VA-TIA	0 (0.0)	1 (1.9)	1.00
Previous psychiatric disease	3 (7.7)	4 (7,7)	1.00
BMI>35	1 (2.6)	1 (1.9)	1.00
Renal Insufficiency	1 (2.6)	0 (0.0)	NA
COPD	2 (5.1)	2 (3.9)	1.00
Previous cancer	2 (5.1)	4 (7.7)	0.70
Previous hematological disease	4 (10.3)	2 (3.9)	0.40
Pathology			
AML	37 (94.9)	46 (88.5)	0.50
MDS	2 (5.1)	6 (11.5)	
AML-t	7 (18.0)	3 (5.8)	0.09
Neutropenia at entry	17 (43.6)	15 (28.9)	0.14
Neurological involvement	3 (7.7)	0 (0.0)	0.08
Hyperleukocytosis	11 (28.2)	14 (26.9)	0.89
Karyotype			
Good prognosis	6 (15.4)	14 (27.5)	0.15
Intermediate prognosis	17 (43.6)	25 (49.0)	
Poor prognosis	16 (41.0)	12 (23.5)	
Complete remission obtained	35 (89.7)	46 (88.5)	1.00
Remedial at D15	8 (20.5)	6 (11.5)	0.24
Remedial at D40	1 (2.6)	2 (3.9)	1.00
Allo-transplant	16 (41.0)	23 (45.1)	0.70
Neutropenia duration PNN <0.5	28.7 (8-81)	32.2 (12-182)	0.42
Neutropenia duration PNN <0.1	19.8 (4-49)	23.3 (9-76)	0.16
Cause of death			
Infection	3 (11.5)	3 (12.5)	NA
Relapse	13 (50.0)	11 (45.8)	
Toxicity	5 (19.2)	8 (33.3)	
GVH	2 (7.7)	1 (4.2)	
Other	3 (11.5)	1 (4.2)	
Hospitalization prior to unit admission			
Yes	10 (25.6)	25 (48.1)	0.03

Abbreviations: NA: Not applicable; BMI: Body mass index; AML: Acute myeloid leukemia; MDS: Myelodysplastic syndrome

Table 2: Infectious characteristics of the population

	Fluconazole (*n*=39)	Posaconazole (*n*=52)	*p*
Duration of prophylaxis	14.2 (2-41)	14,8 (3-42)	0.74
Gut decontamination	25 (64,1)	7 (13.5)	<0,0001
Delay for fever (days)	5.1 (0-14)	4.5 (0-19)	0.60
ATB empiric	39 (100.0)	51 (98.1)	1.00
ATB received			
Imipenem	37 (94.9)	2 (3.9)	<0.0001
Piperacilline-tazo	2 (5.1)	49 (96.1)	
Bi-therapy	6 (15.4)	9 (17.1)	0.81
Nb of ATB received	2.8 (1-5)	2,6 (0-5)	0.38
Nb documented infections			
Bacteremia	14 (34.2)	10 (19.2)	0.10
Pneumonia	5 (12.8)	10 (19.2)	0.41
Infection urinary	1 (2.6)	8 (15.4)	0.07
Sinusitis	1 (2.6)	0 (0.0)	0.43
Infection skin	3 (7.7)	9 (17.0)	0.19
Other	8 (20.0)	3 (5.8)	0.05
No documentation	16 (41.0)	22 (42.3)	1.00
No infection	0 (0.0)	2 (3.9)	0.50
Nb documented infections/patient	0.82 (0-3)	0.77 (0-3)	0.76
Microbe identified			
GNB	3 (7.5)	9 (16.7)	0.23
MRSA	0 (0.0)	1 (1.9)	1.00
MSSA	2 (5.1)	1 (1.9)	0.57
S. coagulase negative	11 (28.2)	10 (19.2)	0.31
Other cocci	4 (10.3)	3 (5.8)	0.46
Other	6 (15.4)	0 (0.0)	0.01
AFE	33 (84.6)	40 (76.9)	0.36
AFE			
Caspofungin	32 (82.1)	1 (1.9)	<0.0001
Liposomal amphotericin B	1 (2.6)	39 (75.0)	
None	6 (15.4)	12 (23.1)	
Delay return of fever	8.1 (3-29)	9,3 (2-34)	0.46
Duration of AFE	14.8 (2-49)	14.6 (1-43)	0.92
Patients treated with voriconazole	15 (38.5)	17 (32.7)	0.57
Delay in return of fever	23.0 (10-40)	17.8 (3-51)	0.24
Pulmonary CT	19 (48.7)	23 (44.2)	0.67
Results			
Negative	5 (26.3)	8 (34.8)	0.55
Non-specific	7 (36.8)	10 (43.5)	
Compatible IPA	7 (36.8)	5 (21.7)	
Fibroscopy BAL	6 (15.4)	7 (13.5)	0.80
IFI			
IFI proven/probable	6 (66.7)	5 (62.5)	1.00
IFI possible	3 (33.3)	3 (37.5)	
Type IFI			
Candidosis	2 (5.1)	0 (0.0)	0.18
Aspergillosis	7 (18.0)	8 (15.4)	0.74
Aspergillosis			
Proven/probable	4 (57.1)	5 (62.5)	1.00
Possible	3 (42.9)	3 (37.5)	

Abbreviations: NA: Not Applicable; CT: Computed tomography; IPA: Invasive pulmonary aspergillosis; BAL: Broncho-alveolar lavage; IFI: Invasive fungal infection

copy with BAL. These examinations added further support to suspicion of a fungal infection in three cases (two examinations were positive for filaments and onebronchoalveolar lavage fluid was very positive upon antigen testing), but these three cases were already classified as probable IPA prior to BAL. Eighteen percent of patients (n = 16) had a positive *Aspergillus* antigen test during their hospitalization. The delay to positive tests was 19.8 days in the posaconazole group versus 14.9 days in the fluconazole group (p = NS). Of the patients with positive antigen results, serology for *Aspergillus* was positive in only 25% of the cases.

Fungal infection characteristics

Proven or probable IFI was found in 11 (12%) of the patients. Taking into account the possible IFIs, the number was 17 (19%), with the difference between the two groups being non-significant. In the fluconazole group, two candidemias were identified (one due to *Candida glabrat*a and the other one due to *Candida krusei*). Histological evidence of the three cases identified as IPA was obtained *a posteriori* (one cerebral biopsy and one pulmonary biopsy positive for filaments with species identification, and one cutaneous biopsy positive for *Aspergillus terreus*). Thirty-six percent of the patients who received remedial chemotherapy on Day 15 developed an IPA (proven, probable, or possible) versus 13% (n = 11) of patients who did not receive additional chemotherapy on Day 15 (p = 0.05). Furthermore, 9 out of 19 patients (47%) who had a poor karyotype prognosis developed an IPA versus 2 out of 18 with a good prognosis and 4 out of 39 with an intermediate prognosis (p = 0.027).

Survival outcomes

Global survival at 3 years was estimated to be 35%. The median survival time for patients without IFI was 27.6 months versus 12 months for patients with an IFI (p = 0.07) [Figure 1A]. No difference in survival as a function of the prophylaxis type used was identified [Figure 1B]. Causes of death were similar in the two groups, with 49% (n = 25) of deaths related to disease relapse. No death was directly imputable to IFI. In the univariate analysis, the estimated risk of death in the presence of an IFI was 1.8 [95% confidence interval (CI): 0.9–3.6]. However, in the multivariate analysis, the only significant factors identified were the karyotypes of poor prognosis and intermediate prognosis [Tables 3 and 4]. Of note, two deaths were reported before the 28th day, both under fluconazole and due to toxicity, in the first 100 days.

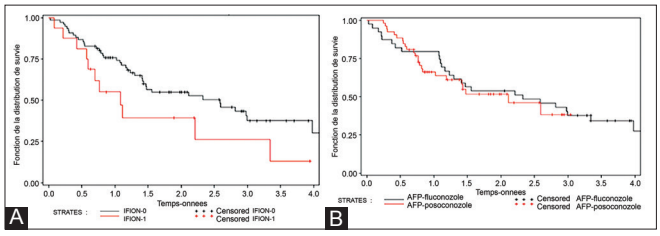

Figure 1: (A) Global survival curve stratified by IFI; (B) global survival curve stratified by prophylaxis type

Mycological colonization

Buccal swaps

Compared to the posaconazole group, we noted a larger proportion of patients colonized with non-*albicans Candida* at study entry in the fluconazole group, a difference which widened at study exit (15% vs 21% for fluconazole; 6% vs 4% for posaconazole). Half of the non-*albicans Candida* isolates were identified as *C. glabrata*. At consolidation, all samples were negative in the posaconazole group as compared with only 77% (n = 14) in the fluconazole group (p = 0.016).

Stool cultures

The microbial types identified in stool cultures are shown in Figure 2A and B. The number of positive cultures in the two groups was identical at study entry [15 positive samples in the fluconazole group vs 14 in the posaconazole group (p = 0.17)] and study exit [19 vs 20 samples (p = 0.22)] [Table 5]. The number of positive samples at entry (n = 29) and exit (n = 39) was statistically different (p = 0.003). The percentage of non-*albicans Can-*

Table 3: Univariate analysis of factors influencing global survival

Univariate analysis	OS	
	HR (95% CI)	p
Hyperleukocytosis	1.13 (0.6-2.09)	0.7
Karyotype		
Poor prognosis	7.7 (2.2-25.9)	0.001
Intermediate	4.9 (1.4-16.3)	0.008
Remedial at D15	1.6 (0.8-3.4)	0.2
Allo-transplant	0.9 (0.5-1.7)	0.9
Empirical ATB	0.9 (0.5-1.7)	0.8
PPI	0.9 (0.5-1.6)	0.7
Presence of IFI	1.8 (0.9-3.6)	0.07

Abbreviations: OS: Overall survival; HR: Hazard ratio; CI: Confidence interval; ATB: Antibiotics; PPI: Proton pomp inhibitor; IFI: Invasive fungal infection

Table 4: Multivariate analysis

Multivariate analysis	OS	
	HR (95% CI)	p
Karyotype		
Poor prognosis	6.7 (1.9-23.7)	0.003
Intermediate	4.9 (1.5-16.5)	0.009
Presence of IFI	1.2 (0.5-3)	0.7

Abbreviations: OS: Overall survival; HR: Hazard ratio; CI: Confidence interval; IFI: Invasive fungal infection

Table 5: Cross table of microbes pre-post, fluconazole versus posaconazole

	Post sortie						
	AL	NA	*C. albicans*	*C. glabrata*	*C. krusei*	Negative	Total
Fluconazole							
Pre							
Other yeast	0	0	0	0	0	1	1
C. albicans	0	1	3	3	1	1	9
C. glabrata	0	0	1	2	0	1	4
C. krusei	0	0	0	0	1	0	1
Negative	0	2	5	0	0	17	24
Total	0	3	9	5	2	20	39
Posaconazole							
Pre							
Other yeast	0	0	0	0	0	1	1
Other NA	0	0	0	0	0	1	1
C. albicans	1	0	4	0	0	5	10
C. glabrata	0	0	0	2	0	0	2
Negative	5	0	8	0	0	25	38
Total	6	0	12	2	0	32	52

Abbreviations: NA: Not applicable; AL: Applicable

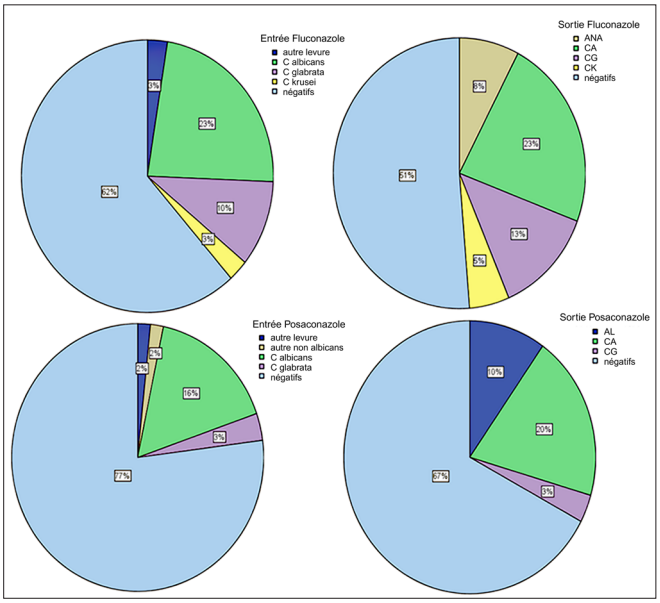

Figure 2: (A) Microbe type in stools pre-post in fluconazole group; (B) microbe type in stools pre-post in posaconazole group

dida progressed between entry and exit, increasing from 13% ($n = 5$) to 24% ($n = 10$). In the posaconazole group, 77% of samples ($n = 38$) were negative versus 67% ($n = 32$) at study exit. Six colonizations with *Saccharomyces cerevisiae* were observed during hospitalization. Finally, at consolidation, 33% of the positive samples in the fluconazole group were *C. glabrata* and 50% of the positive samples in the posaconazole group were *S. cerevisiae*. Two *C. glabrata* (one in each group) had modified their sensitivity to fluconazole at study exit and maintained this change at consolidation.

Cost analysis

The average estimated actualized cost of antifungal therapy during hospitalization was 8973 Euros per patient in the fluconazole group (interval: 342.4–25,348.9€) and 10,062 Euros per patient in the posaconzole group (interval: 1196.5–35,715.9€) ($p = 0.47$). The average cost for empirical therapy was 8395.81€ for the fluconazole group versus 8563.89 for the posaconazole group (p = NA).

DISCUSSION

Until 2007, fluconazole prophylaxis was the treatment of reference for the prevention of IFI in patients receiving hematopoietic stem cell transplantations. This prophylaxis was also often used in other high-risk patients, such as patients with AML receiving induction chemotherapy, even though there was little evidence that the use had an impact on their survival.[35] The study by Cornely *et al.* led to a change in our practice with the label change for posaconazole for use in these patients. The Cornely study demonstrated the superiority of posaconazole over fluconazole and itraconazole in terms of the incidence of IFI (2% vs 8%), with an impact on survival.[7] Despite the homogeneity of our two groups, we did not detect the same difference.

In our study, the incidence of IFI appears to be relatively elevated (12%), notably in the posaconazole group, even though none of the known classic supplementary risks were identified. Despite the availability of modern tools, the diagnosis remains difficult. In order to identify IPA, CT scan should be done when there is the slightest suspicion and should be repeated at regular intervals. The non-specific signs could be compatible with pulmonary aspergillosis and can lead to the diagnosis of possible IPA. The interpretation of CT scans by radiologists sensitized and oriented toward the possibility of aspergillosis could potentially increase the number of possible IPA cases because of the bias. A re-reading of the scans in concert with other members of the team could reduce the risk of this error. BAL is usually performed when the CT scan is abnormal, even in the absence of further suspicion. In our study, the use of BAL did not lead to a change in diagnosis. All the additional mycological evidence was obtained in patients already having a diagnosis of probable IPA due to the presence of host and mycological factors coupled with a compatible CT scan. Even though bronchofiberscopy with BAL can be performed in the majority of patients, it would probably be prudent to limit the procedure to patients with positive radiological signs but with a negative serum antigen result, in which case the BAL analysis could change the diagnosis of an infection from possible to probable. However, the test kit

for the antigen is only validated for serum, and even though a positive result is one of the diagnostic criteria for probable IA (EORTC), no cutoff value has been defined to date.[33] It should be noted, however, that the test can be considered reproducible within the same center.[36,37] The analysis by serum polymerase chain reaction (PCR) in not yet standardized, particularly due to problems with contamination of the material collected. It is not yet part of the diagnostic criteria in actual use.[38] Yet, certain centers use real-time PCR twice per week and begin antifungal therapy as soon as two consecutive positive results are obtained.[39] The working group of the International Society for Human and Animal Mycology is preparing to propose a harmonization of protocols at the European level [European *Aspergillus* PCR initiative (EAPCRI)].[40-42] Finally, the evaluation of the serum β-glucan (present in fungal membranes) could be of global interest, but no evaluation in patients receiving prophylaxis is available.[43]

The absence of difference between the two groups in our study could eventually be explained by the limited sample size. However, there are a number of factors which could influence the metabolism of posaconazole, such as the pharmaceutical interactions or the occasional malabsorption resulting in reduced serum levels. In fact, there are a number of factors favoring the absorption of the molecule (such as a diet rich in fat, liquids with an acidic pH) or limiting its absorption (such as the use of proton-pump inhibitor or metoclopramide, often utilized as an anti-emetic in patients receiving chemotherapy).[44,45] The dose of posaconazole is recommended to reduce this risk.[46] It should be noted that the duration of prophylaxis in this study was relatively short (14 days) in comparison with the Cornely study.[7] This difference is directly related to the rapid change in antifungal toward "empirical" therapy in the case of persistent fever or the occurrence of a new febrile episode. In light of the relatively high incidence of proven/probable IFIs, the value of using empirical therapy can be questioned. In effect, 88% of patients who received voriconazole received prior empirical therapy resulting in three changes of antifungal therapy used during the same hospitalization. In previous studies, empirical and pre-emptive therapies were evaluated without taking into account initial prophylactic therapy, if it was administered. In the Cornely study which evaluated prophylaxis, only 25% of patients required an additional systemic antifungal and then only in of IFI suspicion. Based upon these results, Stam *et al*. developed a predictive model of the average costs of the procedure and ascertained that prophylaxis with posaconazole appeared to be less expensive than with fluconazoleif the reduced incidence of IFI was taken into account.[47] With this in mind, our focus reverts to the other hospitalization costs since no difference was noted between the two groups in terms of prophylaxis cost. This appears to be directly related to the high cost of empirical treatment.

The occurrence of an invasive fungal infection has been described as an independent risk factor for high mortality.[48] No death was attributable to an IFI in this study. We did not note a significant difference with a p value of 0.7. However, this result may have been modulated by the strong impact that the karyotype had in the multivariate analysis and by the restricted sample size. On the other hand, no difference was seen as a function of the type of antifungal protocol used. For risk factors, the influence of remedial chemotherapy on Day 15 was noted, as close to 36% of these patients presented an IFI, as was the influence of a poor prognosis karyotype. It is important to foresee in these populations at high risk for aspergillosis an improvement in earlier diagnosis. It could be discussed whether systematic and regular pulmonary imaging needs to be done from Day 15. Finally, the EORTC actually recommends couple imaging of the sinuses with a pulmonary CT scan in order to detect other locations of invasive aspergillosis, notably in patients having both host and mycological criteria.[32]

In the Cornely study, buccal and stool samples were analyzed once per week. In the two study arms, a decreased incidence of colonization was reported and no selection of species with a decreased sensitivity to azoles was detected. In our study, a significantly increased incidence of stool colonization by non-*albicans Candida* was noted between study entry and exit in patients receiving prophylactic fluconazole, suggesting a selection pressure for this type of prophylaxis.[17-21,49] Conversely, in the posaconazole group, a significantly increased incidence of *S. cerevisiae* was observed at study exit. As far as is known, this emergence is not classically described for this population.[50] It is of interest to note that this "selection pressure" was maintained over time as suggested by the positive results obtained on subsequent hospitalizations, on average 3 weeks later, implying an increased vigilance for the remainder of the patient management.

This study has shortcomings due to its retrospective nature and limited number of patients. Stool and buccal swab cultures have not been validated to assess colonization and efficacy of antifungal therapy; however, they are commonly used in this setting.[51] The retrospective character made collection of data difficult, especially for side effects. We did not observe major toxicities, but it could be explained that patients presenting serious complications were hospitalized in different wards, explaining the absence of data. Our IFI incidence was high, making comparison to other historical series difficult with incidence ranging from 7%[52] with fluconazole prophylaxis to 21% in the absence of prophylaxis.[53] We did not find objective explication for this difference; it could be explained by hygiene, regional and socio-economic disparities. An explanation could be that patients enrolled in this study were likely from a rural region.

Conclusion

The present study does not distinguish any difference-between fluconazole and posaconazole as a primary effective prevention against fungal infections. Nevertheless, one could observe an improvement in sensitivity in the group of patients treated with fluconazole, generating a careful approach. More prospective studies and meta-analyses are warranted.

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