Management of cerebral azole-resistant *A. fumigatus* infection. A role for intraventricular liposomal-amphotericin B?

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Plus est en vous. (Heren van Gruuthuse)

ABSTRACT

Objectives

In the pre-azole era, central nervous system (CNS) infections with *Aspergillus* had a dismal outcome. Survival improved with voriconazole but CNS infections caused by azole-resistant *A. fumigatus* is precluding its use. Intravenous liposomal-amphotericin B (L-AmB) is the preferred treatment option for azole-resistant CNS infections but has suboptimal brain concentrations.

Methods

We describe three patients with biopsy proven CNS aspergillosis where intraventricular L-AmB is added to systemic therapy. 2 patients with azole-resistant and 1 patient with azole-susceptible CNS aspergillosis were treated with intraventricular L-AmB at a dose of 1mg weekly.

Results

We describe 3 patients successfully treated with a combination of intravenous and intraventricular L-AmB. All three patients survived but one patient has serious headache, most likely not related to this treatment.

Conclusions

Intraventricular L-AmB may have a role in the treatment of therapy-refractory CNS aspergillosis when added to systemic therapy.

INTRODUCTION

Few cases of central nervous system (CNS) aspergillosis caused by azole-resistant *Aspergillus fumigatus (ARAF)* have been reported, and almost always with a fatal outcome [1]. Most patients were treated with combination antifungal therapy. Cerebral infections caused by ARAF have almost always a dismal prognosis. Unfortunately, there a no antifungals available that have activity against ARAF and adequately penetrate the brain. Therefore, we added intraventricular liposomal-amphotericin B (L-AmB) to systemic therapy in 3 patients. In this paper, we describe these patients and our clinical experience. All patients provided informed consent.

CASE PRESENTATION

Case 1

An 18-year-old woman with common acute lymphatic leukaemia (ALL) receiving remission induction chemotherapy was diagnosed with a probable invasive pulmonary aspergillosis (IPA). Combination therapy with intravenous (IV) voriconazole and L-AmB (3mg/kg QD) was started. Serum galactomannan was positive (Optical Density (OD) 2.8) and sputum grew an ARAF with the *CYP51A* TR_{34}/L98H mutation. Eight days on therapy, a paresis of the right arm and leg and a right facial nerve paralysis were observed. Brain MRI showed multiple lesions (figure 1A) and a brain biopsy demonstrated hyphae compatible with *Aspergillus* (figure 1C). L-AmB dose was increased to 10 mg/kg and voriconazole (8mg/kg BID) was replaced by posaconazole and dosed at 300mg BID to achieve serum trough concentrations >3mg/L with the hope of achieving therapeutic brain tissue levels. Posaconazole trough concentrations of 5.2 and 6.0 mg/L were documented. Follow-up MRI 15 days after the initiation of therapy showed increased perilesional oedema. During the following 5 months the patient was treated with oral posaconazole 300 mg BID with IV L-AmB daily at 5mg/kg. Six months after diagnosis posaconazole was stopped as cerebral lesions and perilesional oedema had decreased and the arm and leg paresis and facial nerve palsy had improved. Chemotherapy was reinitiated and another 3 months later L-AmB was discontinued. At that time the lesions on brain MRI had decreased in size but not disappeared completely. Unfortunately, six months later the patient was admitted for an epileptic seizure. MRI showed increase in size and oedema around 1 of the 7 lesions. Combination treatment with L-AmB IV (5mg/kg QD) and posaconazole (300mg BID) was reinitiated and was now combined with intraventricular weekly administration of L-AmB (1mg/week). The patient could be discharged with outpatient therapy with L-AmB IV, posaconazole orally and once weekly intraventricular L-AmB using an Ommaya reservoir that was placed for this purpose. The

intraventricular L-AmB therapy was well-tolerated and continued for 4 months. The MRI remained essentially unchanged in these 4 months at that time, the patient was able to walk and cycle independently but has unilateral hand motor dysfunction as the only sequela.

Case 2

A 13-year-old patient with iron overload due to multiple transfusions for beta-thalassaemia was diagnosed with a probable invasive pulmonary and multifocal cerebral aspergillosis 8 months after allogeneic stem cell transplantation (figure 1B). An ARAF was cultured from BAL fluid. Galactomannan in CSF was positive (OD 1.3). Voriconazole (8mg/kg BID IV) and L-AmB (6 mg/kg) were started. Ten days later an epileptic seizure occurred. MRI showed increasing size of the brain lesions and again an ARAF grew from a brain biopsy (table 1). An Ommaya reservoir was placed for the intraventricular administration of L-AmB as well as caspofungin (for details on dosing see table 1). Also intravenous caspofungin (70mg QD) and flucytosine 25mg/kg QID mg) were initiated. With this intervention, the patient improved and lesions decreased in size. Weekly intraventricular administration of L-AmB was continued for 10 weeks and intraventricular caspofungin for 6 months. Systemic therapy with flucytosine, L-AmB and caspofungin was discontinued after 2, 6 and 6 months respectively. No further improvement of the remaining brain lesions was observed after 6 months.

Several years later the patient developed disabling headache. On imaging the skull and dura mater diameter had thickened significantly. A dura biopsy did not lead to a conclusive diagnosis. At last follow-up 9 years post-allogeneic transplant the complains of severe headaches had disappeared but spasticity, occasional epileptic seizures and frontal lobe syndrome have led to severe disability.

Case 3

A 15-year-old girl with common ALL developed aphasia 23 days after chemotherapy initiation. MRI showed 1 lesion in the left frontal and 1 in the temporal lobe. A chest CT showed nodular lesions. BAL sampling was performed. Galactomannan (OD 4.5) was positive and voriconazole-susceptible *A. fumigatus* was cultured (voriconazole MIC 0.5mg/L). Treatment with voriconazole (4mg/kg) and L-AmB (5 mg/kg) was initiated and L-AmB stopped on day 16 when voriconazole drug levels were therapeutic. Despite voriconazole serum levels between 3 and 12 mg/L, a follow-up MRI 3 weeks into therapy showed that lesions had increased in size. Intravenous L-AmB was reinitiated and weekly intraventricular administration of L-AmB 1 mg was started via a Rickham reservoir while voriconazole was continued as well. Follow-up brain MRI's and lung CT at 1 and 2 months into this therapy showed decreasing size of the brain lesions and no increasing lung lesions. A lung biopsy confirmed an invasive aspergillosis infection. Eventually, without

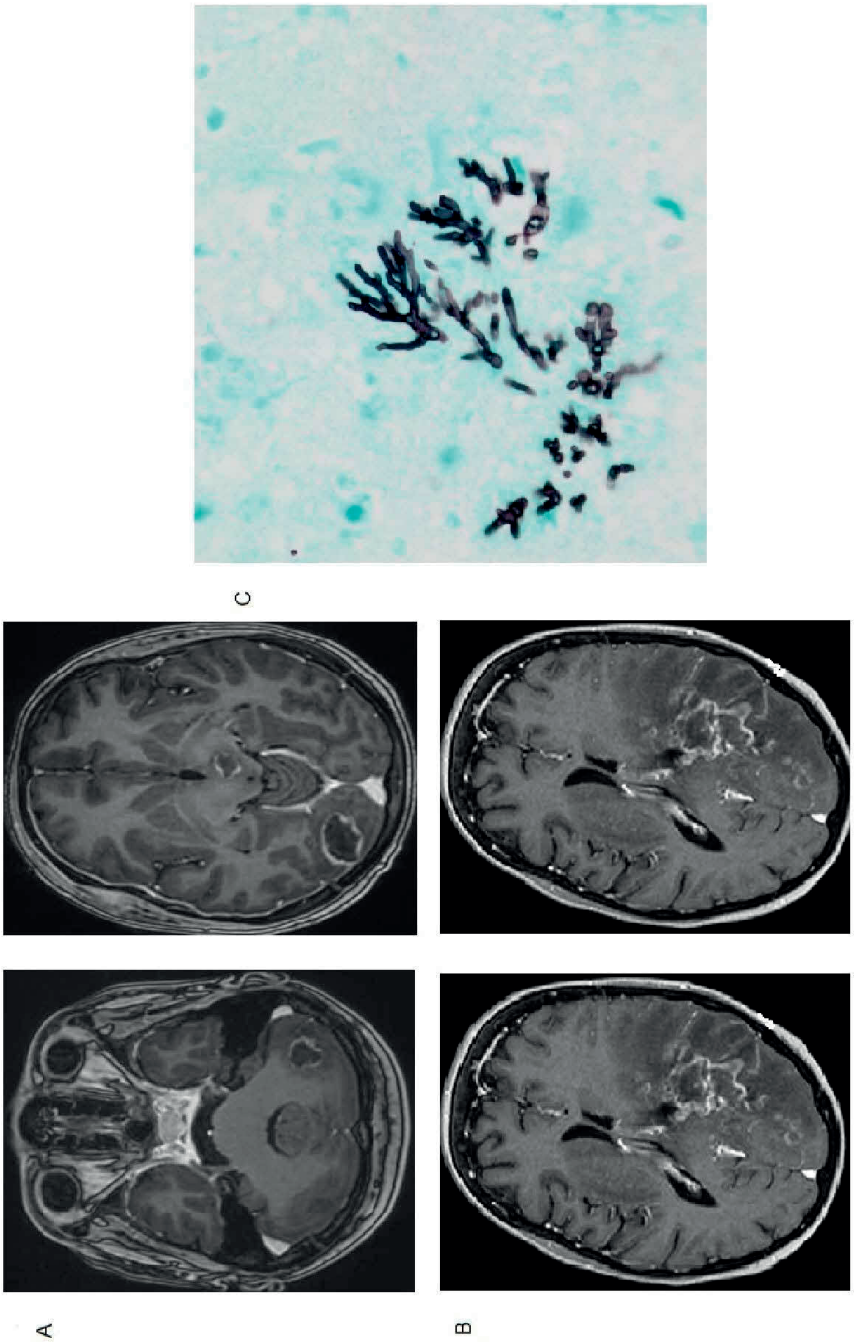

Figure 1: Patient 1, panel A: and patient 2, panel B: MRI showing multiple nodular lesions. Panel C. Brain biopsy shows septated hyphae with dichotomous branching at sharp angles using a gomori methenamine silver stain.

Factors	Patient 1	Patient 2	Patient 3
Sex, Age	F, 18	F, 16	F, 15
Underlying disease	ALL	Thalassemia, allogeneic SCT	ALL
Classification IPA (EORTC/MSG)	Biopsy proven	Biopsy proven	Biopsy proven (brain and Lung)
Culture positive sample	Sputum, brain biopsy	BAL, brain biopsy	BAL
MIC Voriconazole	8	16	0.5
MIC Posaconazole	2	0.5	0.063
MIC Itraconazole	>16	16	0.25
MIC Isavuconazole			0.5
GM value blood	1.3	0.9	0.4
GM value CSF	0.5	1.30	0.5
GM value BAL	2.8	0.36	4.5
Biopsy brain	Positive	Positive	Positive
Treatment regimen (day after diagnosis)	1. Voriconazole + L-AmB IV (3 mg/kg) (d0-d5)	1. L-Amb IV + voriconazole (6 mg/kg) (d0-d10)	1. Voriconazole + L-AmB IV (d0-d10)
	2. Posaconazole + L-AmB IV (10mg/kg) (d5-d26)	2. L-AmB IV + Caspofungin IV (d10-d13)	2. Voriconazole (d11-d20)
	3. Posaconazole + L-AmB IV (3mg/kg) + IT L-AmB* (d26-d191)	3. L-AmB IV + Caspofungin IV/IT$^{\circ}$ (d13-d24)	3. Voriconazole + L-AmB 3mg/kg IV/ weekly IT (d21-d109)
	4. L-AmB 5 mg/kg 3x/ week (d191-d251)	4. L-AmB IV and IT* + Caspofungin IV and IT$^{\circ}$ (d24-d32)	4. Voriconazole (d110-d149)
	5. Treatment re-initiation L-AmB IV + IT *(d386-d515)	5. L-AmB IV and IT* + Caspofungin IV and IT$^{\circ}$ + Flucytosin IV (d32-d97)	5. Voriconazole + L-AmB 3mg/kg IV/ weekly IT* (d149-d176)
		6. L-AmB IV + Caspofungin IV and IT$^{\circ}$ + Flucytosin IV (d97-d201)	6. Isavuconazole + L-AmB 3mg/kg IV and weekly IT (d176-d227)
			7. Isavuconazole + L-AmB weekly IT* (d228-d348)
			8. Isavuconazole (d349-XXX)

Table 1: Clinical and epidemiological characteristics of patients with cerebral azole-resistant invasive aspergillosis

Abbreviations: BAL=bronchoalveolar lavage, CSF=cerebrospinal fluid, F=Female, IPA=invasive pulmonary aspergillosis, EORTC/MSG= European Organization for Research and Treatment of Cancer/ Invasive Fungal Infections Cooperative Group and the National Institute of Allergy and Infectious Diseases Mycoses Study Group [EORTC/MSG] host factor, F=Female, IPA=invasive pulmonary aspergillosis, IT=intrathecal/intraventricular, IV=intravenous, L-AmB=Liposomal Amphotericin B, MIC=minimal inhibitory concentration. *Intraventricular/intrathecal L-AmB was given at a dose of 1mg weekly $^{\circ}$Caspofungin IT 1 mg QD for 2 weeks1mg 3x/week for 1 month and 3x/week 0.5 mg thereafter

a change in therapy all infections sites improved, as well as the neurological disabilities of the patient. After 3 months of combination therapy, a step-down to voriconazole monotherapy was made. During the following 6 weeks voriconazole levels were suboptimal (range 0.3-2.6mg/L) and unfortunately, 6 weeks later dysarthria developed and an MRI showed that lesions had increased in size. A brain biopsy confirmed the *Aspergillus* infection of the brain. IV and intraventricular L-AmB was reinitiated. IV administration of L-AmB was stopped after two months but intraventricular continued for 8 months while voriconazole was switched to isavuconazole for liver enzyme elevations. Eventually follow-up imaging of lungs and brain showed a good response to therapy. Patient is doing well and has been successfully treated for ALL. During ALL therapy, patient is receiving isavuconazole as secondary antifungal prophylaxis.

DISCUSSION

We describe 2 cases of ARAF and 1 case with azole-susceptible CNS aspergillosis treated with intraventricular L-AmB. Brain infections with *Aspergillus* have a high mortality and survivors are left with at least some neurological deficit [2]. Although voriconazole improved the chances of survival, ARAF now turns back the clock to the amphotericin-B era.

Over the last 10 years, azole-resistance has become an important emerging problem and is associated with a very high mortality [3-6]. When voriconazole resistance is documented in a patient infected with a cerebral *Aspergillus* infection, treatment becomes very difficult. Indeed, few other systemic antifungal agents have been shown to penetrate the brain. Furthermore, the therapeutic effect of new drugs is still unknown [7, 8]. Pharmacokinetic and pharmacodynamic animal data suggest that compared with other formulations of amphotericin-B, L-AmB results in the highest brain tissue concentrations of amphotericin-B and it was effective as therapy in a mouse model of candida encephalitis [9]. Therefore, it is regarded as the preferred second-line therapy for cerebral fungal infections but should, at least initially, be dosed at 5 to 10 mg/kg to achieve therapeutic brain tissue concentrations quickly [1, 10]. For azole-resistant CNS infection, L-AmB can be combined with a second drug but itraconazole, posaconazole and echinocandins do not lead to adequate drug concentrations in CSF or brain tissue with standard dosing regimens [1]. Furthermore, it seems that combination therapy does not lead to synergistic treatment effect *in vitro* against azole-resistant *A. fumigatus* isolates [11]. To improve the CSF and brain penetration higher systemic exposure may be aimed for to subsequently reach higher CSF and brain concentrations that can exert a pharmacological effect even in the setting where pathogen susceptibility is reduced. Furthermore, a damaged blood-brain barrier, which is the case in patients with an angio-invasive *Aspergillus* infection will likely improve the penetration of selected

drugs. With this in mind, we combined posaconazole with L-AmB in the first case [12]. Ultimately, we decided to administer L-AmB directly into the cerebrospinal fluid space as well. The administration was well tolerated with no subjective side effects. Contrary to the first and third case, we observed long-term complications in the second case. We argue that these side effects are probably the result of chronic inflammation and scarring during and after the infection rather than L-AmB or caspofungin mediated toxicity although we cannot exclude with certainty that local combination of drug therapy contributed to these side effects.

Current guidelines do not recommend the use of intraventricular administration of antifungals due to the risk of important adverse events (e.g. chemical meningitis, seizures) [13]. Historically, intrathecal/intraventricular administration of conventional AmB deoxycholate has been and is still being used to treat patients with coccidioidal meningitis. The side effects of intrathecal/intraventricular administration of AmB deoxycholate make it difficult to use and only low doses of typically 0.1 mg are used after which the dose is slowly increased up to 1.0 mg [14]. The reported side effects of AmB deoxycholate, led us to opt for intraventricular L-AmB instead. Based on a theoretical total CSF volume of approximately 100-150 mL in our patients, the administration of 1mg of L-AmB would result in a peak CSF concentration of L-AmB of 10 microgram/mL- which is comparable to the peak plasma concentrations after systemic administration by Groll *et al* [9]. Distribution kinetics as well as clearance mechanism were unknown so we had no knowledge on possible accumulation, hence we started with a presumed safe dose. In case 1 we tried to measure L-AmB in retrospect on left-over CSF fluid but no L-AmB could be detected (limit of detection 0.5 mg/L). In hindsight we argue that the clearance of L-AmB is much more rapid than initially expected. This is explained by the fact that 500 ml of CSF is produced and reabsorbed each day and helps clearing L-AmB. Both the dose and frequency of once weekly intraventricular administration of 1 mg L-AmB might thus be suboptimal and a higher dose as well as a more frequent administration may be preferred for future patients. Intrathecal/intraventricular L-AmB at a higher dose (10mg per administration) for seven consecutive days was shown to be well tolerated in 18 patients with cryptococcal meningitis [15]. Although the exact role of intrathecal/intraventricular L-AmB remains to be defined in patients with an *Aspergillus* infection of the CNS, we propose to initiate intrathecal/intraventricular L-AmB as soon as voriconazole resistance is documented at a dose of 5mg and preferably twice weekly. If available, L-AmB CSF concentration monitoring may guide dosing after the first dose.

Finally, whether local therapy needs to be given in conjunction with systemic therapy is unknown. The benefit of combination therapy may be a more favourable ratio of plasma/brain concentrations and perhaps a longer detainment of adequate CSF/brain concentrations.

Case series like ours have several limitations. In particular, all 3 patients received systemic treatment as well. Therefore, the exact contribution of the intraventricular L-AmB administration cannot be defined. However, it is very unlikely that prospective clinical studies will ever be performed to find the best possible treatment option for very rare infections like CNS aspergillosis. Therefore, treatment should be based on in vitro and animal data and eventually the experience described in case reports and case series can be helpful as well.

In conclusion, 3 patients with a CNS infection with *A. fumigatus* were treated with combination antifungal therapy that included intraventricular L-AmB. All 3 survived but one patient was left with severe sequalae.

Funding

This study was performed as part of our routine work.

Ethical approval

Not required.

Transparency decleration

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