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Liposomal amphotericin B in the treatment of fungal infections in neutropenic patients: a single-centre experience of 133 episodes in 116 patients

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Summary. Liposomal amphotericin B (AmBisome) was used for suspected or confirmed fungal infection complicating 133 neutropenic episodes in 116 patients not tolerating, or not responding to, conventional amphotericin. Adverse effects were infrequent and no significant renal impairment resulted. Acute reactions occurred in five patients, reversible hepatic dysfunction in 23, and hypernatraemia in 17. The putative mycosis resolved with AmBisome treatment in 81 episodes (61%) and progressed with fatal outcome in 25 (19%), but the diagnosis was equivocal in most, and in 27 episodes (20%) evidence indicating nonfungal pathogenesis emerged. Treatment efficacy is, however, evaluable in those with proven aspergillosis. 13/17 patients with confirmed invasive aspergillosis responded to AmBisome (77%), conventional amphotericin having failed in 11. Treatment was successfully discontinued when the neutrophil count was $< 1 \times 10^9/l$ in eight responders (61%). In four further patients treated for suspected aspergillosis, disseminated infection was documented at post-mortem, but the true incidence is unknown. This analysis confirms that AmBisome is well tolerated and effective against invasive mycoses in neutropenic patients, and may salvage patients when conventional amphotericin proves excessively toxic or ineffective.

Keywords: fungal infection, neutropenia, aspergillosis, liposomal amphotericin.

Fungal organisms are increasingly recognized as serious and potentially fatal pathogens in the immunosuppressed host, but mycotic disease remains difficult to identify accurately, and to treat effectively. The manifestations of invasive fungal infection in profoundly immunocompromised subjects are nonspecific, often indistinguishable clinically from bacterial, viral and protozoal infections, radiation pneumonitis and graft-versus-host disease (Clift, 1984; Chopra *et al*, 1991; Rubin & Greene, 1988; Singer *et al*, 1979). Moreover, the causative organisms are infrequently isolated despite invasive investigation (Bustamante & Wade, 1990; Greenman *et al*, 1975; Miale *et al*, 1987; Stover *et al*, 1984) and the pathogens identified may result from colonization or contamination. *In vitro* sensitivities to available antimycotic drugs may not predict clinical response, and resistance may emerge during treatment (DeGregorio *et al*, 1982; Powderly *et al*, 1988). Furthermore, whereas the adverse effects of available antifungal antibiotics are well documented, their efficacy in eradicating systemic fungal infections in the severely immunocompromised patient has not been established (Denning & Stevens, 1990; Fisher *et al*, 1981; Hay, 1991).

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Patients treated for haematological malignancies are particularly susceptible to fungal infection due to prolonged periods of profound immunosuppression (Gerson *et al*, 1984), the most commonly identified pathogens being aspergillus, candida, mucormycosis and cryptococcus (Denning, 1991). Mycotic infection is frequently invasive and carries a significant mortality; Denning & Stevens (1993) describe mortality rates of greater than 90% in neutropenic patients with proven aspergillosis despite therapy. Amphotericin B is the only proven effective antifungal active in deep-seated infections due to these organisms (Bodey & Vartivarian, 1989) but it is toxic.

Adverse effects from amphotericin B are common, and include reactions such as pyrexia, rigours, phlebitis, myalgias, malaise, nausea and vomiting, and bronchospasm, and more serious dose-limiting organ damage (Warnock, 1991). Nephrotoxicity is dose dependent and common, and ranges from mild diabetes insipidus and renal tubular defects to acute renal failure. Reversible cardiac dilatation, hepatic

dysfunction and marrow suppression are recognized complications (Stein & Tolle, 1983; Warnock, 1991). Interstitial pneumonitis and the respiratory distress syndrome have been attributed to amphotericin B, but may equally result from the pulmonary infection (Singer *et al*, 1979).

In an attempt to circumvent toxicity, several different formulations of amphotericin B have been developed, the most promising of which involves incorporation of the drug into small liposomes. Phase II and III trials suggest that liposomal amphotericin, whilst causing minimal adverse effects, achieves relative drug targeting to the reticuloendothelial system, including the pulmonary alveolar macrophages, hepatic Kuppfer cells and tissue histiocytes, where fungal infection predominates (Meunier *et al*, 1991; Tollemar *et al*, 1990). It is possible that liposomal amphotericin is more effective than the conventional drug (Lopez-Berenstein *et al*, 1995; Ringden *et al*, 1991).

We have used liposomal amphotericin B in neutropenic patients with proven or suspected invasive fungal infection, who had not tolerated, or not responded to, therapeutic doses of conventional amphotericin B, who were deemed of high enough risk of succumbing to fungal infection to merit the use of a new drug. A retrospective analysis of the results is presented. The first 20 patients were reported by Chopra *et al* (1991).

MATERIALS AND METHODS

Patients. Between January 1991 and May 1993, more than 750 episodes of neutropenia occurred in patients with haematological conditions undergoing care at University College Hospital. 375 bone marrow transplants were performed, and the neutropenia resulted from intensive chemotherapy administered for acute leukaemia or lymphoma, or progression of the disorder in the remainder. Systemic antifungal therapy was administered for suspected or proven mycosis in approximately two-thirds of these neutropenic episodes, and in 133 liposomal amphotericin was substituted for the conventional formulation. Details of the 116 patients treated with the liposomal drug are given in Table I.

Table I. Patient details.

Patients	Total 116
Age	Median 31 years (range 7–65)
Sex	Female 33 (28%); male 83 (72%)
Diagnosis	
ALL 18 (16%)	AML 32 (28%); NHL 25 (22%);
HD 22 (19%)	MM 7 (6%); CGL 10 (9%)
SAA 2 (2%)	
Treatment (133 episodes)	
BMT 54 (38 auto, 16 allo)	Chemotherapy 76; ATG, steroids 3

ALL: acute lymphoblastic leukaemia; AML: acute myeloblastic leukaemia; HD: Hodgkin's disease; NHL: non-Hodgkin's lymphoma; CGL: chronic granulomatous leukaemia; SAA: severe aplastic anaemia; BMT: bone marrow transplantation; ATG: antithymocyte globulin.

Supportive care and antifungal treatment. Patients undergoing allogeneic marrow transplantation were nursed in isolation rooms with positive-pressure filtered air. All other patients were cared for in single rooms with simple reverse-barrier precautions but no filtered air. Antifungal prophylaxis was instituted at the onset of neutropenia, using nystatin suspension (Nystan, Squibb) and amphotericin pastilles (Fungilin, Squibb), and in four patients, while building work was undertaken on the ward, nebulized amphotericin B. Broad-spectrum antibiotic therapy was initiated immediately pyrexias greater than 38°C occurred, after cultures had been taken. Antifungal treatment was started if pyrexia persisted despite antibiotics for greater than 72 h, or for proven or strongly suspected fungal infection. Although the manifestations of fungal infection are nonspecific, several constellations of clinical symptoms and signs were thought suggestive of fungal infection. Table II shows the numbers of AmBisome recipients with these signs.

Table II. Signs suggesting fungal infection.

(a)	Bilateral interstitial shadowing (BIS) on CXRay, and fever, cough (often nonproductive), pleuritic pain	56 (48%)
(b)	Pulmonary nodules (some cavitating)	32 (28%)
(c)	Multiple hepatic and splenic nodules	9 (8%)
(d)	Stomatitis and oesophagitis with plaques	9 (8%)
(e)	Pneumonia unresponsive to antibiotics	6 (5%)
(f)	Pyrexia unresponsive to antibiotics	14 (12%)

Microbiological confirmation of infection with aspergillus was obtained in 15 cases from clinical specimens (see Table III), primarily from the respiratory tract. In four patients widespread invasive aspergillosis was found on post-mortem examination, despite intensive treatment with amphotericin during the final illness. In two patients a presumptive diagnosis of aspergillosis was supported by serially increasing titres of aspergillus antigens (Pasteurex). Thus only 21 (18%) of the AmBisome-treated patients had firm evidence of aspergillus infection, and in four the diagnosis was only established post-mortem. In 15/21 candida was also cultured from throat swabs and faecal samples.

Microbiological evidence of candidosis was obtained in 56 patients (42%). In 30 of these, throat swabs and stool cultures were positive on at least two occasions; and in 23 candida was also isolated from additional specimens (Table III). A putative candidal aetiology for sterile endocarditis or multifocal hepatosplenic abscesses was augmented by increasing agglutinin titres in three cases.

In only a small proportion of isolates was accurate speciation attempted, and antifungal drug sensitivities were unobtainable. Fluconazole (Diflucan, Pfizer) 100–200 mg/d orally was first-line therapy for gastrointestinal candidosis. Low-dose intravenous amphotericin B (0·25 mg/kg/d) was used for azole-resistant infection, and for patients unable to tolerate oral medication. For documented or suspected deep-seated fungal infection, amphotericin B was administered in gradually increasing doses as tolerance permitted, to a daily

Table III. Confirmation of fungal infection.

Aspergillus		Candida	
Sputum isolate	8	From throat and faeces only	30
From lung biopsy	1	Plus sputum	16
From liver biopsy	2	Plus bronchoalveolar lavage	3
From pleural effusion	1	Plus tracheal aspirate	2
From bronchoalveolar lavage	2	Plus blood	2
From Hickman line swab	1		
Increasing agglutinins	2	Increasing agglutinins	3
At post-mortem	4		
Total	21 (18%)	Total	56 (48%)

dose of 1·2 mg/kg, until toxicity intervened, or the signs resolved. Liposomal amphotericin B (AmBisome, Vestar) was substituted for the conventional formulation if adverse effects prevented, or infection progressed despite, adequate treatment. In special circumstances, AmBisome was used as initial therapy.

Liposomal amphotericin B has been used on 133 occasions, in 116 patients. In 99 episodes (74%) conventional amphotericin B had been administered for a median of 8 d (range 1–30) and dose 420 mg (80–1900), prior to the liposomal drug. AmBisome was substituted because of deteriorating renal function (increase in serum creatinine to at least twice baseline levels) in 59 episodes (60%), and for other severe adverse effects in seven. Clinical and radiological evidence of progression of infection despite conventional amphotericin B (median dose 660 mg, 340–1470), prompted AmBisome rescue in 32. The switch was made on two occasions to allow recipients to continue antifungal treatment as outpatients. In 34 treatment episodes (26%) AmBisome was the initial antifungal therapy. In 16 this was due to

Table IV. Outcome in patients with documented aspergillosis.

Pt	Age (yr)	Sex	Diag.	Chemo.	Signs	m+c	Day	ConA (mg)	Why	Day	LipA (mg)	Resp.	CR	NC
1	22	F	ALL	Intens.	BIS pl. eff.	Pl. eff.	18	490	Renal	12	700	Compl.	Y	<1
2	30	M	NHL	ABMT	BIS cough	Sputum	9	530	Prog.	10	830	Compl.	Y	>1
3	19	M	AA	ALG	BIS pl. pain	Sputum	15	850	Prog.	7	800	Compl.	N	<1
4	35	F	AML	ABMT	L+S nodules	L bx	6	750	Prog.	96	4290	Compl.	Y	>1
5	45	F	AML	ABMT	BIS pl. eff.	BAL	6	420	Prog.	9	760	Compl.	Y	<1
6	7	M	AML	Consol.	L+S nodules	L bx	8	620	Prog.	96	4260	Compl.	Y	>1
7	46	M	AML	Induct.	Pulm. nod. sinus	Sputum	30	1470	Prog.	7	660	G rec.	N	<1
8	72	M	AML	Induct.	Pulm. nodules	Aggl.	0	0	Renal	56	4290	Compl.	Y	>1
9	54	M	AML	Reind.	Pulm. nodules	Sputum	1	80	Renal	16	3200	VG	N	<1
10	34	M	ALL	Induct.	BIS cough	Hline	6	280	Renal	8	1030	Compl.	Y	<1
11	52	M	MM	BMT	BIS cough	Sputum	0	0	Renal	19	2400	Compl.	N	>1
12	19	M	NHL	BMT	BIS cough	Sputum	4	125	Renal	8	800	Compl.	Y	<1
13	34	M	ALL	Induct.	Pulm. nodules	Sputum	7	560	Prog.	11	3300	VG	Y	<1
14	16	M	ALL	Induct.	BIS cough	P bx	7	490	Prog.	6	800	d MUF	N	<1
15	32	F	HD	ABMT	BIS tracheitis	Sputum	6	380	Renal	9	1350	d RF	N	<1
16	67	M	AML	Induct.	BIS cough	BAL	20	1200	Prog.	5	650	d RF		<1
17	63	M	AML	Induct.	BIS cough	Sputum	0	0	Renal	9	900	d MUF		<1
18	27	M	HD	ABMT	BIS pl. eff.	PM	25	1230	Renal	7	520	d MUF		<1
19	35	M	ALL	Induct.	Pulm. nodules	PM	9	650	Prog.	8	1600	d MUF		<1
20	20	M	ALL	Induct.	BIS cough	PM	6	430	Prog.	16	4025	d RF	N	<1
21	31	M	CGL	BMT	BIS cough	PM	17	810	Renal	8	700	d MUF	N	<1

Pt: patient; Diag.: diagnosis; Chemo: chemotherapy; Induct.: induction; Reind.: reinduction; Consol.: consolidation; Intens.: intensification; ALG: antilymphocyte globulin; ABMT, BMT: autologous or allogeneic transplantation; BIS: bilateral interstitial shadowing; Pl. eff: pleural effusion; Pulm.: pulmonary; L, S: liver, spleen; sinus: sinus mass; m + c: source of proof; BAL: bronchoalveolar lavage; bx: biopsy; Hline: Hickman line; Aggl: increasing agglutinins; ConA: conventional amphotericin; mg: total dose; Why: reason for change; Prog.: progression of infection; Renal: nephrotoxicity; lipA: AmBisomse; compl., VG, G: complete or partial resolution; d: died; RF, MUF: respiratory, multiorgan failure; CR: complete remission; NC: neutrophil count at discontinuation.

recurrence of the infection previously requiring AmBisome during subsequent neutropenia; in eight pre-existing renal impairment, in five lack of central venous access, and in four to permit outpatient therapy.

The median duration of liposomal amphotericin treatment was 12 d, with a range of 2–96 d, and the median dose administered 1684 mg (180–10440).

RESULTS

Safety of liposomal amphotericin B

AmBisome was well tolerated, even at the dose of 5 mg/kg/d. Five patients (4%) experienced chills, rigours or nausea during infusion, which could be prevented with hydrocortisone and chlorpheniramine. No thrombophlebitis was documented in patients in whom AmBisome was infused via a peripheral line.

No clinically significant deterioration in serum urea or creatinine resulted from liposomal amphotericin use. Rapid recovery of these parameters to normal occurred following AmBisome substitution in all patients with renal toxicity from the conventional formulation, but polyuria and solute loss due to tubular damage was slow to resolve, and electrolyte replacement was required during subsequent AmBisome therapy. Accurate documentation of renal tubular function with *ab initio* liposomal amphotericin was

Table V. Outcome in patients with signs suggesting aspergillosis.

Pt	Age (yr)	Sex	Diag.	Chemo.	Signs	Investigation	Day	ConA (mg)	Why	Day	LipA (mg)	Result	NC
1	15	M	ALL	Consol.	L+S nodules	Liver bx	15	960	Prog.	20	2000	Compl.	>1
2	36	M	NHL	ABMT	BIS pl. pain	CXR BAL	15	490	Renal	7	1000	Compl.	>1
3	46	M	AML	Induct.	Pulm. nodules	CXR CTscan	4	280	Renal	7	730	VG	>1
4	26	F	AML	Induct.	Pulm. nodules	CXR CTscan	18	405	Renal	8	960	Compl.	>1
5	52	M	AML	Induct.	Pulm. nodules	CTscan BAL	12	580	Renal	11	1000	Compl.	>1
6	39	M	HD	ABMT	Pulm. nodules	CTscan	7	460	Outpt.	9	1200	Compl.	>1
7	51	F	AML	ABMT	BIS pl. pain	CTscan BAL	6	530	Prog.	41	2750	Compl.	>1
8	50	M	AML	Consol.	Pulm. nodules	CXR CTscan	6	660	Prog.	20	4000	Compl.	>1
9	36	M	NHL	ABMT	Pulm. nodules	CXR BAL	8	280	Renal	17	1940	Compl.	>1
10	17	F	ALL	Intens.	Pulm. nodules	CXR CTscan	0	0	Outpt.	10	1200	VG	>1
11	17	M	AML	Induct.	Pulm. nodules	CXR	4	320	Renal	38	5600	VG	<1
12	35	F	ALL	Reind.	Pulm. nodules	CXR CTscan	6	580	Prog.	23	3850	Compl.	>1
13	16	M	AML	Induct.	Pulm. nodules	CXR CTscan	8	750	Prog.	14	2600	VG	<1
14	15	M	ALL	Intens.	Pulm. nodules	CXR CTscan	6	500	Prog.	30	9000	Compl.	>1
15	54	F	AML	Consol.	Pulm. nodules	CTscan BAL	7	580	Prog.	12	1920	Compl.	<1
16	55	M	CGL	Immun.	Pulm. nodules	CXR	0	0	Renal	12	600	Compl.	>1
17	28	M	HD	ABMT	Pulm. nodules	CTscan BAL	6	480	Renal	5	750	VG	>1
18	52	F	ALL	Intens.	Pulm. nodules	CXR CTscan	3	260	Renal	10	1500	VG	>1
19	34	M	ALL	Induct.	Pulm. nodules	CTscan BAL	7	550	Prog.	10	3000	G sur.	>1
20	30	M	HD	Cytred.	L+S nodules	CTscan BAL	20	940	Outpt.	14	1400	IsHD	>1
21	32	F	NHL	Cytred.	L+S nodules	Liver bx	8	320	Renal	7	700	IsNHL	>1
22	52	M	ALL	Consol.	Pulm. nodules	CXR	10	420	Renal	4	600	d RF	<1
23	29	M	HD	Cytred	Pulm. nodules	BAL +bx	0	0	Renal	5	500	d HD	>1
24	21	M	AML	Induct.	BIS pl. pain	CXR BAL	6	230	Intol.	16	3850	d RF	>1
25	24	M	HD	ABMT	Pulm. nodules	BAL +bx	0	0	p iv	14	2100	p HD	>1
26	27	F	NHL	ABMT	Pulm. nodules	CXR BAL	6	600	Renal	5	500	d RF	<1
27	58	M	MM	Induct.	Pulm. nodules	CTscan BAL	4	280	Renal	7	1050	p TB	>1
28	27	M	AML	ABMT	L+S nodules	CTscan	15	930	Prog.	32	1550	dRF	<1
29	18	F	HD	ABMT	BIS pl. pain	CTscan BAL	?	?	Renal	8	560	d RF	<1
30	31	F	HD	Cytred.	Pulm. nodules	CTscan BAL	8	450	Renal	28	2800	p HD	>1
31	43	F	NHL	ABMT	Pulm. nodules	CTscan BAL	12	360	Renal	9	450	p NHL	>1
32	44	M	MM	ABMT	BIS pl. pain	BAL lung bx	3	150	Renal	33	3800	pfibr	>1
33	46	M	NHL	ABMT	BIS pl. pain	CXR	9	395	Renal	13	1650	d RF	<1
34	47	F	NHL	ABMT	Pulm. nodules	CXR CTscan	0	0	Renal	11	1100	p NHL	>1
35	31	F	HD	ABMT	Pulm. nodules	CXR CTscan	6	520	Prog.	20	1700	d HD	>1
36	55	F	NHL	ABMT	Pulm. nodules	CTscan BAL	8	680	Prog.	12	1200	d RF	>1

Pt: patient; Diag.: diagnosis; Chemo.: chemotherapy; Induct.: induction; Reind.: reinduction; Consol.: consolidation; Cytred.: cytoreduction; Intens.: intensification; ABMT: autologous transplantation; BIS: bilateral interstitial shadowing; pl. eff: pleural effusion; pulm, p: pulmonary; L, S: liver, spleen; CXR: chest radiography; CTscan: computerized tomography; BAL: bronchoalveolar lavage; bx: biopsy; ConA: conventional amphotericin; mg: total dose; Why: reason for change; Prog.: progression; p iv: no central venous access; LipA: AmBisome; compl, VG, G: complete or partial resolution; sur.: surgical resection; d: died; RF: respiratory failure; fib.: fibrosis; NC: neutrophil count at discontinuation.

precluded by concurrent diuretic and nephrotoxic medication in the inpatients, and infrequent monitoring in outpatients, but no outpatient required supplements. Hypernatraemia (Na > 150 mmol/l) developed in 17 patients during AmBisome therapy (15%). Haemofiltration was instituted for sodium concentrations exceeding 170 mmol/l in two patients, one of whom did not survive. A causal link between liposomal amphotericin and the hypernatraemia in these critically ill patients has not been substantiated.

Abnormalities of hepatic function possibly attributable to liposomal amphotericin developed during 23 (17%) episodes, in two cases severe enough to warrant discontinuation of the drug. The median peak level of aspartate transaminase was 103 IU/l (range 58–510), alkaline phosphatase 582 IU/l (302–1362) and bilirubin 55 μmol/l (15–310). The abnormalities resolved on drug withdrawal.

Outcome of treatment with liposomal amphotericin

(a) *Proven aspergillus.* Although attempts to confirm the diagnosis frequently included invasive procedures, aspergillosis was only proven in 21 patients with signs indicative of the infection, albeit in four only at post-mortem. 13 of these 21 (62%) patients (77% of those diagnosed ante-mortem) obtained complete or excellent partial resolution of the clinical and radiological signs of infection (Table IV). In eight the liposomal amphotericin was discontinued when the neutrophil count was below 1×10^9/l, without recrudescence of the infection. 11 of the 21 received AmBisome for deteriorating infection despite adequate doses of conventional amphotericin B, and seven of these (64%) responded to liposomal amphotericin. Complete haematological remission was confirmed in nine responders on recovery from chemotherapy. Three were found to have persistent disease, and two suffered recurrent aspergillosis following subsequent chemotherapy. Of the eight nonresponders, four had refractory malignancy, and four died before assessment was possible.

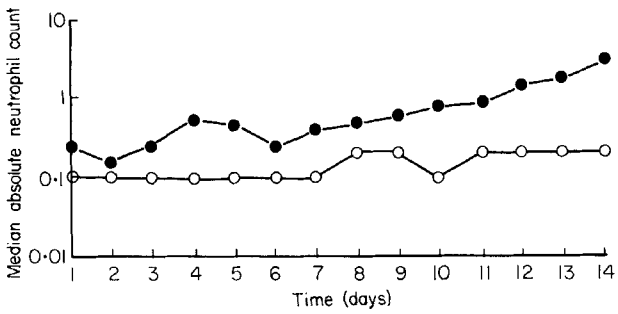

Fig 1. Median neutrophil count during the initial AmBisome treatment period: (●) responders ($n = 13$); (○) nonresponders ($n = 8$).

Fig 1 depicts the median absolute neutrophil count (ANC) for the first 14 d of AmBisome therapy in responders and nonresponders. In patients benefiting from AmBisome the initial median ANC was $0{\cdot}25 \times 10^9$/l, rising to $1{\cdot}2 \times 10^9$/l at 14 d. The median ANC for nonresponders was $0{\cdot}1 \times 10^9$/l when liposomal amphotericin was started, and did not exceed $0{\cdot}2 \times 10^9$/l in the subsequent fortnight ($P = 0{\cdot}003$). However, not all responders had rising granulocyte counts; in three the ANC was consistently less than $0{\cdot}5 \times 10^9$/l for the first 2 weeks of therapy. In two AmBisome was discontinued when the ANC was $< 0{\cdot}5 \times 10^9$/l, without ill effect.

(b) *Patients with suspected aspergillosis.* Table V depicts the results of AmBisome treatment in 36 patients with strongly suspected but unproven invasive aspergillosis. 19 (53%) attained complete or good partial eradication of the signs of infection, which had progressed in eight (42%) despite conventional amphotericin. In four responders AmBisome was safely discontinued despite ongoing immunosuppression (ANC $< 1 \times 10^9$/l in three, high-dose steroids in one). One responder later underwent pulmonary resection for cavitatory sequestra. Fig 2(a) demonstrates the radiographic findings, typical of this group, which were thought to represent fungal infection. Fig 2(b) shows the improvement

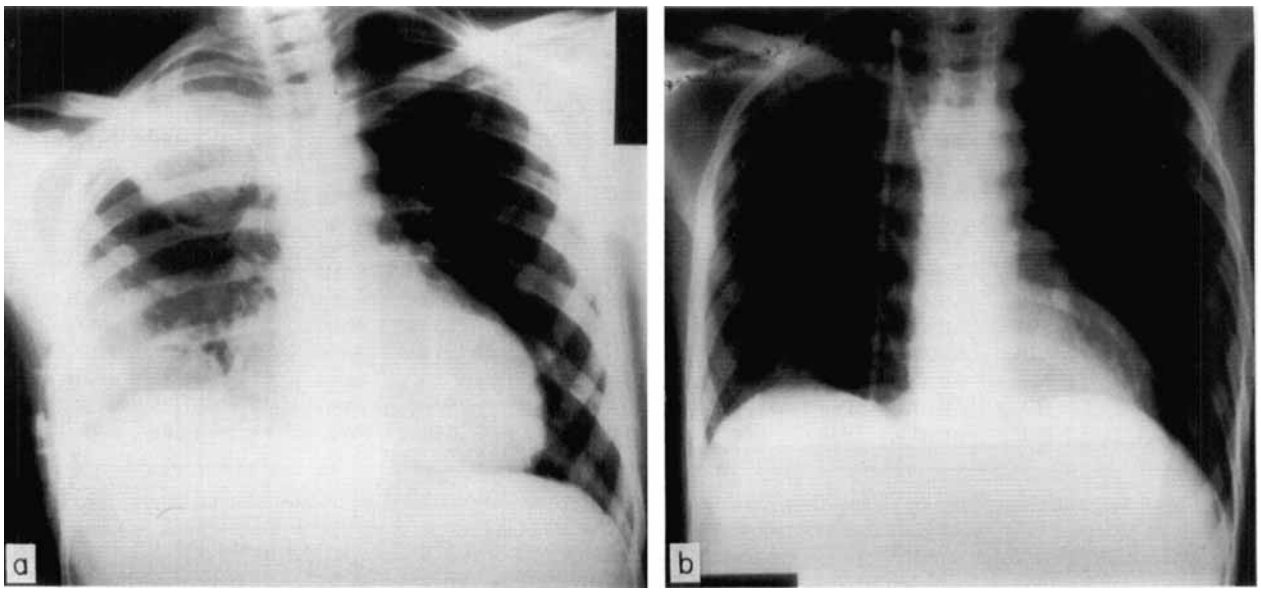

Fig 2. (a) Radiographic appearance typical of patients with suspected aspergillosis; (b) the response after treatment with liposomal amphotericin.

after 14 d treatment with liposomal amphotericin. 11 of the 19 patients benefiting from AmBisome received further intensive chemotherapy, in six for refractory malignancy; 10 developed recurrent signs. Palliative care was offered to two other responders with resistant malignancy. Complete remission was achieved in 11/19 (58%).

Fourteen patients (39%) did not improve with AmBisome therapy: in five the pneumonic process progressed and proved fatal, and one succumbed to liver failure. Post-mortem was refused in all six, but four had evidence of refractory malignancy. In eight nonresponders the clinical signs attributed to aspergillosis were ultimately shown to be due to resistant lymphoma, tuberculosis or lung fibrosis.

(c) *Patients with candidosis.* Candidal septicaemia was documented in two of the 116 neutropenic patients receiving AmBisome, and proved fatal in one. In the remaining 54 subjects the relevance of the isolate to the systemic disease is not clear. Mucosal candidosis did not always respond to liposomal amphotericin B, possibly due to amphotericin resistance or decreased bioavailability of the liposomally bound drug at these sites. The clinical and microbiological features of oropharyngeal candidosis persisted in three of the five patients given the drug *ab initio* for this indication, and monilia was cultured from throat swabs and faecal samples during AmBisome treatment in seven of the 15 with isolates of candida in addition to aspergillosis. Similarly, in the 48 other patients with culture-positive monilia, candida was isolated in 11 despite at least 7 d of therapy.

(d) *Outcome of all patients.* Overall, in 81/133 treatment episodes a successful outcome was achieved (61%). 13 patients had proven aspergillosis, and 39 candida. In 25 episodes, judged as treatment failures (19%), the patient deteriorated and died despite AmBisome therapy. Eight of these patients had microbiological evidence of aspergillosis, and nine candida. In 27 episodes (20%) the signs prompting antifungal therapy were found to have an alternative aetiology (Table VI).

Table VI. Pathogenesis in the 27 nonmycosis patients.

Visceral deposits of lymphoma	14
Pulmonary	9
Hepatosplenic	3
Meningeal	1
Pneumonitis due to nonfungal organisms	12
Cytomegalovirus	5
Streptococcus	2
Tuberculosis	2
Pseudomonas	2
Pneumocystis	1
Pulmonary haemorrhage	1

DISCUSSION

The use of liposomal amphotericin B in this large series of patients with presumed or proven invasive fungal infection confirms the safety data from previous smaller studies (Chopra *et al*, 1991; Lopez-Berenstein *et al*, 1985; Meunier *et al*, 1991; Tollemar *et al*, 1990). There was a low incidence of immediate reactions, such as fever, chills and rigours, making the drug far more acceptable to patients and staff than conventional amphotericin. This, combined with the short infusion times and absence of thrombophlebitis when peripheral veins are used, has allowed outpatient treatment with liposomal amphotericin B in selected cases. Of particular relevance is the lack of nephrotoxicity which frequently limits the use of conventional amphotericin B. Not only did renal function not deteriorate with AmBisome, but in patients with renal impairment induced by conventional amphotericin, improvement invariably occurred after the liposomal form was substituted. Two potential toxicities from liposomal amphotericin B were observed. Firstly there was a high incidence of hepatocellular dysfunction in AmBisome recipients, although it must be noted that such abnormalities are frequent in this group of severely ill patients and a causal relationship has not been proven. The hepatocellular toxicity was reversible when liposomal amphotericin was discontinued, but this coincided with recovery from infection and the effects of chemotherapy. Secondly, there was a surprisingly high incidence of hypernatraemia in these patients which was fatal in one case. It is difficult to ascribe this with confidence to the liposomal amphotericin in these patients. All had received prior conventional amphotericin B, and had suffered some degree of tubular damage, which resolves slowly. Several had abnormal fluid compartmentalization and were given diuretics, and all received other potentially nephrotoxic agents concurrently. Furthermore, there is no obvious explanation as to why hypernatraemia should be caused by liposomal amphotericin B, which contains negligible amounts of sodium.

Efficacy of antifungal agents in severely immunocompromised patients is notoriously difficult to assess. Overall in this series 61% of the patients treated with AmBisome recovered, but it is inevitable that several probably never actually had fungal infection or had colonization unrelated to the systemic illness. The most important efficacy data in this paper thus relates to the 21 patients with proven aspergillus infection. 13 of the 17 patients (75%) in whom the aspergillus was identified during life, responded to liposomal amphotericin, which compares very favourably with conventional amphotericin B in this situation, where mortality rates in excess of 90% are reported (Denning, 1990; Wilson & Denning, 1993). In some patients, aspergillus cannot be isolated during life despite considerable efforts, and a definite diagnosis can only be made at post-mortem. Four such patients are included in this series, but it must be acknowledged that post-mortems were not performed on all patients. Furthermore, some patients with aspergillus in whom positive identification was never made were probably cured by the liposomal amphotericin. It is thus not possible to identify the precise incidence of aspergillus infections in our patients and give an accurate overall value for treatment efficacy.

It has been suggested previously that eradication of invasive fungal infection only occurs once neutrophil recovery has begun, even with amphotericin B therapy (Chopra *et al*, 1991; Denning, 1991). In the studies reported by Denning & Stevens (1990) and Fisher *et al* (1981) there were high

mortality rates in persistently neutropenic patients with aspergillosis, and resolution of fungal infections was associated with the attainment of complete remission and neutrophil recovery. This analysis is broadly in agreement, in that responding patients tended to have higher neutrophil levels which rose over the following 2 weeks, when compared to the non-responding patients. It should be noted, however, that the median neutrophil count at the time liposomal amphotericin was started in the responders was only $0{\cdot}25 \times 10^9$/l, which would still be considered severe neutropenia. Furthermore, in three patients, response to liposomal amphotericin occurred without the neutrophil count rising above $0{\cdot}2 \times 10^9$/l, and in two cases AmBisome was stopped without recrudescence of infection when the neutrophil count was still $<0{\cdot}5 \times 10^9$/l. Liposomal amphotericin may therefore be of value in patients without a rising neutrophil count.

A major issue is whether liposomal amphotericin B is not just an effective agent against fungal infections but is more effective than the conventional form of the drug. The data presented here offer some encouragement to that view, in that seven of the patients with proven aspergillosis who responded to liposomal amphotericin B had failed to respond to the conventional form of the drug immediately before. However, with the passage of time, there is an increased likelihood of neutrophil recovery and the relative efficacy of the two forms of amphotericin can only be resolved by a randomized trial. This is logistically extremely difficult to execute because of the difficulty in obtaining proof of aspergillus infection and because therapy must be instigated on suspicion before such proof is obtained.

In conclusion, liposomal amphotericin B is an effective agent for the treatment of aspergillus infections, and is far less toxic than the conventional form of this drug. It may be less efficacious in mucosal candida infections. Although its high cost may restrict its use, we believe it should be considered in any immunocompromised patient with a high suspicion of severe fungal infection who is intolerant of conventional amphotericin or who fails to respond to this treatment.

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