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Dose-Dependent Antifungal Activity and Nephrotoxicity of Amphotericin B Colloidal Dispersion in Experimental Pulmonary Aspergillosis

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We investigated the safety and efficacy of amphotericin B colloidal dispersion (ABCD) for the treatment of invasive pulmonary aspergillosis in persistently granulocytopenic rabbits. Treatment groups included ABCD in dosages of 1, 5, and 10 mg/kg/day intravenously or conventional desoxycholate amphotericin B (DAmB) at 1 mg/kg/day intravenously. Antifungal activity was directly related to increasing dosage of ABCD as determined by the concentration of *Aspergillus fumigatus* organisms in lungs and the frequency of hemorrhagic pulmonary lesions. At 5 and 10 mg/kg/day, there was a significant reduction in the tissue burden of *A. fumigatus* as measured by percent culture-positive lobes and CFU per gram of tissue ($P \leq 0.001$), whereas at 1 mg/kg/day the tissue burden of *A. fumigatus* was not significantly different from that in untreated controls. Microbiological clearance was significantly greater at 1 mg of DAmB per kg per day than at 1 mg of ABCD per kg per day ($P \leq 0.001$). There was no difference in microbiological clearance of bronchoalveolar lavage fluid among the treatment groups as measured by CFU per milliliter. As determined by survival, ABCD at 5.0 mg/kg/day was more effective than DAmB at 1.0 mg/kg/day and ABCD at 10 mg/kg/day. ABCD at 10 mg/kg/day was more nephrotoxic than the lower dosages of ABCD and resulted in higher mortality. Impairment of glomerular filtration developed as a direct function increasing the ABCD dosage ($r = 0.77$; $P < 0.001$). In summary, this study found dose-dependent antifungal activity and nephrotoxicity of ABCD against invasive pulmonary aspergillosis in persistently granulocytopenic rabbits and showed that the optimal dosage of ABCD for antifungal activity and safety was 5 mg/kg/day.

Invasive pulmonary aspergillosis is an important cause of morbidity and mortality in patients with chemotherapy-induced granulocytopenia (6, 9, 16–18, 21, 23). Current treatment of invasive pulmonary aspergillosis with desoxycholate amphotericin B (DAmB) is often unsuccessful and may be complicated by nephrotoxicity (3, 16). Novel antifungal agents that offer improved therapeutic efficacy and reduced toxicity are critically needed for this infection. During the past decade, different lipid formulations of amphotericin B have been developed to increase its antifungal activity and decrease its toxicity (1–4, 7, 10, 12, 14, 15, 20).

Recently, a novel formulation, amphotericin B colloidal dispersion (ABCD), has been developed by using the specific interaction of amphotericin B with sterols (10, 11). Amphotericin B and sodium cholesteryl sulfate at an equimolar ratio form a thermodynamically stable discoidal complex of uniform size. Initial in vivo studies demonstrate that the therapeutic index of the drug is significantly improved (four- to sixfold) as a direct result of the unique physical properties of this colloidal system (5, 11, 19). This drug-lipid complex does not cause hemolysis and shows low binding to plasma lipoproteins in vitro. Although ABCD is intended for treatment of invasive aspergillosis, little is known about its antifungal activity and nephrotoxicity in treatment of invasive pulmonary aspergillosis in persistently granulocytopenic hosts, the most common setting for this form of *Aspergillus* infection.

We therefore investigated ABCD in the treatment of experimental invasive primary pulmonary aspergillosis in persistently granulocytopenic rabbits. A panel of complementary therapeutic indices of antifungal therapy are used in this model to facilitate a precise understanding of the potential efficacy of ABCD in the treatment of pulmonary aspergillosis in persistently granulocytopenic hosts.

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MATERIALS AND METHODS

Animals. Female New Zealand White rabbits weighing 2.5 to 3.5 kg were used throughout these experiments. Animals were individually housed and provided food and water ad libitum, following National Institutes of Health guidelines on care and use of laboratory animals. Silastic venous catheters were surgically placed under sterile operative conditions (22). This chronic silastic central venous catheterization provided nontraumatic venous access for induction, maintenance, and support of persistent granulocytopenia in the rabbits.

Immunosuppression and supportive care. Cytosine arabinoside (kindly provided by Upjohn, Kalamazoo, Mich.) at 525 mg/m^2 was administered intravenously on days 1 through 5 and on days 8 and 9 to produce profound and persistent granulocytopenia ($\leq$100/μl) monitored by serial leukocyte counts (Cell Counter Analyzer; Coulter Electronics, Inc., Hialeah, Fla.) and slide differentials. Methylprednisolone (Upjohn), 5 mg/kg, was administered on days 1 through 3 to inhibit macrophage conidiacidal activity. Rabbits were closely monitored and supported throughout granulocytopenia. Granulocyte counts were maintained below 500/μl and usually below 100/μl from day 5 onward. Ceftazidime (Glaxo, Research Triangle Park, N.C.) at 75 mg/kg intravenously twice daily, gentamicin (Baxter Health Care Corp., Deerfield, Ill.) at 5 mg/kg intravenously daily, and vancomycin (Eli Lilly, Indianap-

olis, Ind.) at 15 mg/kg intravenously daily, were administered from day 4 onward to prevent the emergence of opportunistic bacterial infections during granulocytopenia. Serial samples of blood were drawn for monitoring renal and hepatic function. Chemical determinations included serum creatinine, urea nitrogen, aspartate aminotransferase, alanine aminotransferase, and bilirubin (Ani Lytics, Gaithersburg, Md.).

Preparation and administration of *Aspergillus* inoculum. *Aspergillus fumigatus* conidia from a patient with histologically proven pulmonary aspergillosis were used in all experiments. The organism was grown on potato dextrose agar, maintained frozen at −70°C, thawed, subplated onto several more potato dextrose agar slants, incubated for 24 to 48 h, and kept at room temperature until its use 5 days later. Under a laminar-flow hood, 7 ml of 0.025% Tween saline solution was instilled into each potato dextrose agar slant, and *A. fumigatus* conidia were harvested from the surface by gentle scraping with a transfer pipette. The suspensions of conidia were centrifuged at 3,000 × *g* for 10 min. The conidia were counted with a hemacytometer, and an inoculum of 6.0×10^8/ml was prepared by resuspension into 0.025% Tween saline solution for inoculation. Each rabbit received 1.5×10^8 conidia via the endotracheal route, suspended in 250 μl, on day 2 of each experiment.

Each rabbit was anesthetized intravenously with 0.8 to 1.0 ml of a 2:1 mixture (by volume) of 100 mg of ketamine (Fort Dodge Labs, Fort Dodge, Iowa) per ml and 20 mg of xylazine (Mobay Corp., Shawnee, Kans.) per ml for analgesia, amnesia, and skeletal muscle relaxation. The anesthetic dosage was adjusted according to body weight in order to achieve similar depths of general anesthesia in all rabbits. Once satisfactory anesthesia was reached, a laryngoscopically directed Flagg 0 straight blade (Welch-Allyn, Skaneateles Falls, N.Y.) was carefully inserted until the vocal cords were clearly visualized. The *A. fumigatus* inoculum was then aspirated by a tuberculin syringe attached to a 5.25-in (ca. 13.3-cm) Teflon catheter (Becton Dickinson, Sandy, Utah), and the inoculum was administered beyond the vocal cords into the trachea under direct visualization. Careful intubation was ensured, and the esophagus was not inoculated. This process was repeated for each rabbit enrolled in the particular trial.

Antifungal therapy. DAmB (Fungizone; Bristol Myers-Squibb, Princeton, N.J.) was reconstituted with distilled water and then diluted with 5% glucose in water to a 1-mg/ml concentration immediately prior to use. Antifungal therapy with 1 mg of DAmB per kg or 1, 5, or 10 mg of ABCD per kg was initiated on day 3, 24 h after inoculation. These compounds were administered daily over 10 consecutive days.

ABCD was reconstituted with sterile water to a concentration of 5 mg/ml. The vial was gently rocked until a yellow opalescent suspension without aggregates was obtained. This solution was injected directly or diluted to a 1-mg/ml solution in 5% glucose in water, depending on the dosage and volume administered. Prior to reconstitution, DAmB and ABCD were stored at 4°C protected from light. Bottles of reconstituted DAmB and ABCD were wrapped in aluminum foil, stored at 4°C, and used within 24 h.

Five treatment groups were studied in 72 rabbits: ABCD at 1, 5, and 10 mg/kg/day, DAmB at 1 mg/kg/day, and untreated infected controls. DAmB and ABCD were infused at 0.4 ml/min intravenously through the central silastic venous catheter. Both compounds were well tolerated, and there was no clinically overt infusion-related toxicity.

Postmortem studies. The rabbits were sacrificed with intravenous pentobarbital. The heart and lungs were resected en bloc. Lung weights were obtained and recorded. Lung weights in experimental aspergillosis reflect the extent of pulmonary hemorrhage and edema related to infection. The lungs were carefully inspected for the presence of any lesions and specifically scored by two observers blinded to the treatment group for hemorrhagic or resolving lesions. Hemorrhagic lesions were defined macroscopically as foci of necrosis and hemorrhage. Histologically, these lesions were characterized by the presence of hyphal angioinvasion, hemorrhagic infarctions, coagulative necrosis, karyorrhexis, and few mononuclear cells. Resolving lesions were defined macroscopically as hyaline to red-gray lesions without necrosis or fresh hemorrhage. Microscopically, resolving lesions were characterized by a dense cellular infiltrate composed of fibroblasts, macrophages, mononuclear cells, and multinucleate giant cells with occasional granuloma formulation. No hyphae are typically observed on hematoxylin-and-eosin-stained specimens. The number of lobes involved with macroscopic lesions was recorded and tabulated as an overall percentage for each rabbit group for comparative analysis. After bronchoalveolar lavage (see below), thin sections of lesions were prepared and preserved in 10% neutral buffered formalin at room temperature for histopathological studies.

Postmortem bronchoalveolar lavage was performed by cannulating the trachea with a 12-ml syringe filled with sterile saline solution. A 10-ml volume of saline was instilled gently into the tracheobronchial tree and then withdrawn. The process was performed three times. Each bronchoalveolar lavage specimen was then placed in cytospin, stained by the Wright technique, and examined for the presence of hyphae and macrophages. Bronchoalveolar lavage fluid (0.1 ml) was cultured on Sabouraud glucose agar.

Following tissue sectioning for any lesions, portions of each of the rabbit's six lobes were weighed and homogenized in sterile normal solution. CFUs were then determined for each lobe by using Sabouraud glucose agar plates incubated at 37°C for 24 h and then placed at room temperature. Colonies were counted at 48 h and recorded as CFU per gram of tissue. The number of lobes that yielded positive cultures for *Aspergillus fumigatus* was also recorded as a percentage for each rabbit.

Statistical analysis. Differences in survival were measured by Kaplan-Meier analysis (13). Differences in proportions were analyzed by chi-square or Fisher's exact test. Differences between means were measured by unpaired Student's *t*-test. The correlation between serum creatinine and dosage of ABCD was determined by Spearman rank correlation. All analyses were two sided. $P \leq 0.05$ was considered significant.

RESULTS

Survival. Survival of rabbits receiving 1 or 5 mg of ABCD per kg per day was significantly greater than that of controls ($P \leq 0.001$) (Fig. 1). There was a trend toward greater survival in the 1- and 5-mg/kg/day treatment groups than in the DAmB-treated control rabbits ($P = 0.09$). The greatest survival was achieved in the treatment group receiving 5 mg of ABCD 5 per kg per day. In comparison with rabbits receiving 1 or 5 mg/kg/day, survival in the group receiving 10 mg of ABCD per kg per day decreased markedly ($P = 0.03$), apparently as a result of nephrotoxicity.

Microbiological clearance. There was a dose-dependent relation of ABCD to microbiological clearance (Table 1). When the dosage was 5 and 10 mg/kg/day, there was a significant reduction in tissue burden of *A. fumigatus* as measured by percent culture-positive lobes and by CFU per gram of tissue ($P \leq 0.001$), whereas when the dosage was 1 mg/kg/day, the tissue burden of *A. fumigatus* was not significantly different from that in untreated controls. Microbiologi-

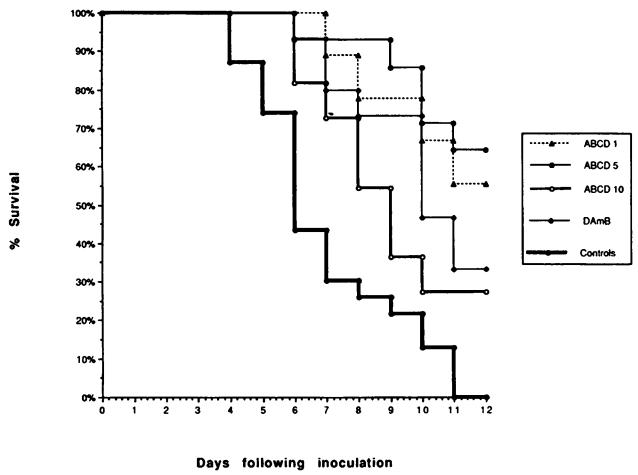

FIG. 1. Survival curve of persistently granulocytopenic rabbits with invasive pulmonary aspergillosis treated with DAmB at 1 mg/kg/day or increasing dosages of ABCD at 1, 5, or 10 mg/kg/day.

cal clearance in DAmB-treated control rabbits was similar to that in the rabbits given 5 or 10 mg of ABCD per kg per day. Microbiological clearance was significantly greater at 1 mg of DAmB per kg per day than at 1 mg of ABCD per kg per day ($P \leq 0.001$).

Cultures of bronchoalveolar lavage fluid were found to be positive only in rabbits with culture-positive lung tissue. Of the 15 animals that received treatment with ABCD or DAmB and still had one or more culture-positive lobes, 3 (20%) yielded positive bronchoalveolar lavage cultures, whereas 8 (36%) of 22 untreated rabbits with positive lung culture had a positive BAL culture.

Tissue injury and pulmonary hemorrhage. Tissue injury due to invasive pulmonary aspergillosis was determined by the lung weights and by the number of lobes with lesions at the autopsy study. There was a dose-dependent relation between the ABCD dose and tissue injury measured by lung weights (Table 1). Rabbits treated with 5 and 10 mg of ABCD per kg per day had significantly greater reduction of lung weights in than did those treated with 1 mg of ABCD per kg per day. The reduction of lung weights in DAmB-treated control rabbits was similar to that in the rabbits treated with 5 and 10 mg of ABCD per kg per day. The reduction in lung weights was significantly greater in rabbits treated with 1 mg of DAmB per kg per day than in those treated with 1 mg of ABCD per kg per day.

There were two patterns of pulmonary lesions: hemorrhagic lesions and resolving lesions. Hemorrhagic lesions were present in most lobes of untreated controls (Table 2). By comparison, in treated animals these lesions were significantly reduced in number by antifungal therapy, particularly in the group treated with 5 mg of ABCD per kg per day ($P \leq 0.001$). Resolving lesions were found only in treated animals.

Nephrotoxicity. Rabbits in all groups receiving amphotericin B had a significant increase in serum creatinine during the course of therapy ($P = 0.05$ to 0.002) as measured by the penultimate serum creatinine in comparison with the baseline serum creatinine value (Fig. 2A). However, there was no significant increase in serum creatinine in untreated controls. There also was a dosage-dependent trend in nephrotoxicity due to ABCD (Fig. 2B). The mean inverse serum creatinine, which is directly proportional to creatinine clearance, decreased with increasing dosage of ABCD ($r = -0.77$, $P < 0.001$; Spearman rank correlation).

DISCUSSION

This study found a dose-dependent antifungal effect and nephrotoxicity of ABCD in a persistently granulocytopenic rabbit model of experimental pulmonary aspergillosis. Among the markers of safety and efficacy studied for dose response in this system were survival, microbiological clearance, pulmonary score, and renal function. Because granulocytopenia is a major risk factor for invasive pulmonary aspergillosis (9, 24), the antifungal activity and toxicity of ABCD were studied by using a persistently granulocytopenic rabbit model of invasive pulmonary aspergillosis. The respiratory route of inoculation was studied to represent the most common route of infection in granulocytopenic patients.

Survival in the different treatment groups is a marker of both antifungal activity and toxicity of the compound. Survival in ABCD-treated rabbits was greatest at a dosage of 5 mg/kg/day and lowest at 10 mg/kg/day. Given the effective microbiological clearance of ABCD at 10 mg/kg/day, the deleterious effect on survival may be due to toxicity of the compound. Normal rabbits have a mean serum creatinine of approximately 0.7 to 0.8 mg/dl. A rapid rise of serum creatinine to 3.0 to 4.0 mg/dl is frequently fatal in rabbits and may account for the greater mortality in the treatment group receiving 10 mg of ABCD per kg per day. Other immunomodulatory mechanisms, such as the induction of tumor necrosis factor alpha, at this high dosage also may be a factor contributing to excess mortality at the highest dosage of ABCD used in this study.

As measured by tissue burden of *A. fumigatus* (log [CFU per gram]), the frequency of culture-positive lobes, and the lung weight as a marker of intrapulmonary hemorrhage, ABCD at 10 mg/kg/day was more active than the lower dosages of 5 and 1 mg/kg/day. Reflecting the greater antifungal activity, there also were trends toward a reduced percentage of animals with hemorrhagic lesions and a greater proportion of resolving lesions at increasing dosages ranging from 1 to 10 mg/kg/day.

Hemorrhagic infarction due to vascular invasion is an important factor in the pathogenesis of invasive pulmonary aspergillosis in pancytopenic patients. Because *Aspergillus* hy-

TABLE 1. Comparative efficacies of ABCD and DAmB in treatment of experimental primary pulmonary aspergillosis

Treatment group and dosage (mg/kg/day)	Survival (%)[a]	% of lobes culture positive[a]	Log CFU/g of lung tissue[a]	Lung wt (g)[a]
ABCD, 10 (n = 11)	27 d,¶,§	4.5 ± 3.2 a,e	0.11 ± 0.56 c,g	12.7 ± 1.7 b,*
ABCD, 5 (n = 14)	64 c,§	9.5 ± 7.1 a,e	0.25 ± 0.81 b,f	14.4 ± 1.4 c,†
ABCD, 1 (n = 9)	56 b,¶	66.7 ± 11.1 e	1.60 ± 1.20 e,f,g	22 ± 3.6 b,*†
DAmB, 1 (n = 15)	33 a	13.3 ± 5.4 a	0.28 ± 0.73 a,e	15.3 ± 1.5 a
Controls (n = 23)	0 a,b,c,d	57.6 ± 5.2 a	1.41 ± 1.45 a,b,c	41.3 ± 3.1 a,b,c

[a] Significance of paired treatment groups by chi-square analysis or unpaired Student's t test: a to d, $P \leq 0.001$ in comparison with untreated controls; e to g, $P \leq 0.001$; †, *, ¶, §, $P \leq 0.05$.

TABLE 2. Distribution of hemorrhagic and resolving lesions in treatment groups of experimental primary pulmonary aspergillosis

Treatment group and dosage (mg/kg/day)	No. (%) of rabbits with lesions	No. (%) of rabbits with hemorrhagic lesions[a]	No. (%) of rabbits with resolving lesions[a]
ABCD, 10 (n = 11)	11 (100)	3 (28) d	8 (72) d
ABCD, 5 (n = 14)	7 (50)	4 (58) c	3 (42) c
ABCD, 1 (n = 9)	8 (88.8)	5 (63) b	3 (37) b
DAmB, 1 (n = 15)	13 (86.6)	6 (46) a	7 (54) a
Controls (n = 23)	19 (82.6)	19 (100) a–d	0 (0) a–d

[a] a to d, $P \leq 0.01$ in comparison with untreated controls.

phae invade tissue, these hemorrhagic lesions may evolve rapidly. High-dosage amphotericin B may, as a result of concentration-dependent killing, permit rapid interdiction of hyphal invasion and prevention of hemorrhagic lesions. Hemorrhagic lesions that have already formed do not remain static. In the absence of further progression due to invasive aspergillosis, these lesions begin to resolve. The histological hallmarks of these resolving lesions are large numbers of pulmonary alveolar macrophages, mononuclear cells, and fibroblasts. These lesions have a distinctive gray hyaline appearance that distinguishes them from the opaque hemorrhagic infarcts. Thus, a correlation may be drawn between the dose response of an antifungal agent and the development of pathophysiologically distinctive pulmonary lesions in invasive pulmonary aspergillosis.

Despite the dose-dependent antifungal effects of ABCD in treatment of experimental invasive pulmonary aspergillosis, the nephrotoxicity of the compound appeared to be a dose-limiting factor. There was a direct relation between increasing dosage and increasing impairment of glomerular filtration as reflected by the inverse creatinine function. Whether there are toxic effects concomitant with nephrotoxicity remains to be further elucidated. For example, increased production of tumor necrosis factor alpha inhibition of thrombopoiesis, impaired coagulation, or increased myelosuppression may further contribute to dose-dependent toxicity.

ABCD was less nephrotoxic on a milligram-per-kilogram basis than was conventional DAmB. For example, administration of ABCD at 1 mg/kg/day to male Sprague-Dawley rats resulted in 3.3- to 3.6-fold lower concentrations in the kidneys on day 14 after treatment, in comparison with those of DAmB; these lower renal levels of ABCD were associated with reduced nephrotoxicity (8). A similar relationship between lower concentrations of ABCD in kidney tissue and reduced nephrotoxicity was also observed in dogs by the same investigators (7). Although high concentrations of ABCD, approximately five times that of DAmB, were found in livers of animals receiving this compound, these elevated concentrations were not associated with an increase in liver toxicity, nor were there increased levels of circulating hepatic transaminases in our rabbits receiving ABCD at dosages of 1 through 10 mg/kg/day.

Conventional treatment with DAmB at 1 mg/kg/day, compared with ABCD at 1 mg/kg/day, was more active in causing microbiological clearance, in reducing the frequency of culture-positive lobes, and in reducing lung weights. Antifungal activity of ABCD comparable to that of DAmB was not achieved until a dosage of 5 mg/kg/day was used. There was a trend toward superior activity of ABCD at 10 mg/kg/day over that of DAmB at 1 mg/kg/day. However, the toxicity of ABCD at 10 mg/kg/day abrogated potential survival benefits of this enhanced antimicrobial activity. That ABCD in equivalent dosages to DAmB had less antifungal activity was also found

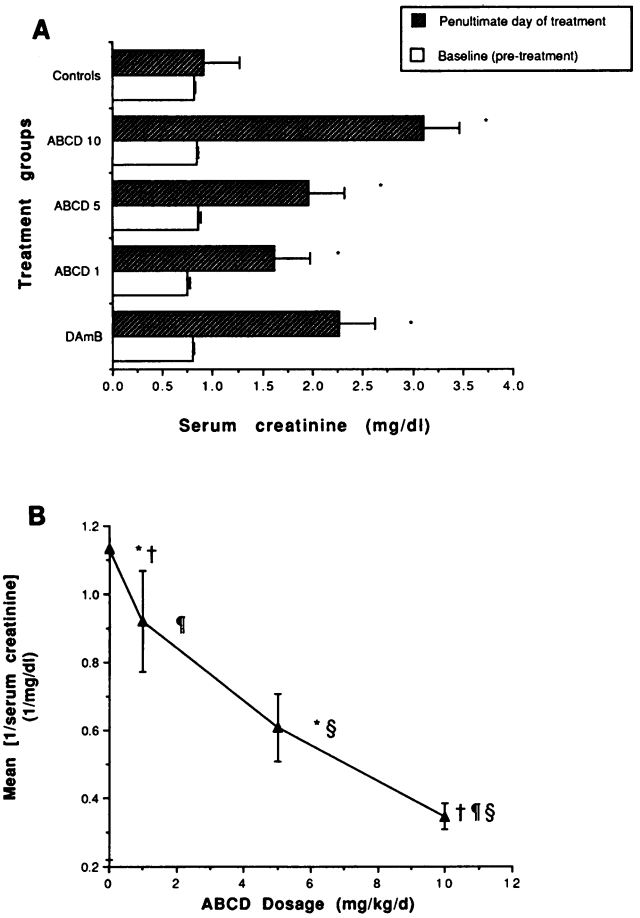

FIG. 2. (A) Changes in serum creatinine in granulocytopenic rabbits with pulmonary aspergillosis treated with ABCD or DAmB. All rabbits in groups receiving ABCD or DAmB had a significant increase in serum creatinine (*, P = 0.05 to 0.002) during the course of therapy as measured by the penultimate serum creatinine in comparison with the baseline serum creatinine. (B) Dosage-dependent trend in nephrotoxicity due to ABCD. The mean inverse of serum creatinine, which is directly proportional to the mean creatinine clearance, decreases with increasing dosage of ABCD ($r = -0.77$; $P < 0.001$); *, §, $P < 0.05$; †, ¶, $P < 0.005$.

by Patterson et al. in an immunocompromised-rabbit model of disseminated aspergillosis (19).

The differences in survival between rabbits treated with ABCD at 5 mg/kg/day versus DAmB at 1 mg/kg/day are likely to be attributable to the combined effects of enhanced rate of tissue clearance, decreased pulmonary injury, and reduced nephrotoxicity in the ABCD group. The differences in survival between these two treatment groups cannot be explained exclusively by the relatively small differences observed in nephrotoxicity. They also cannot be ascribed to postmortem tissue burden, since the concentrations of *A. fumigatus* were similar.

The differences in survival also may be related to the rate of clearance of organisms, since the high-dosage regimen of ABCD at 5 mg/kg/day may have interdicted the infection more rapidly than did the DAmB regimen. Supporting this possible mechanism are the differences in the number of rabbits with pulmonary lesions (7 of 14 [50%] versus 13 of 15 [87%] [P = 0.05, Fisher's exact test] for ABCD at 5 mg/kg/day and DAmB at 1 mg/kg/day, respectively) between the treatment groups.

Pulmonary injury due to vascular invasion and hemorrhagic infarction is a critical factor in the pathogenesis of pulmonary aspergillosis. An antifungal regimen that can more rapidly damage hyphae and halt the progression of vascular invasion will probably reduce pulmonary injury and improve survival. Consistent with these findings are the results of a concurrent study performed during these experiments of serial chest computed tomograms of rabbits for therapeutic monitoring of invasive aspergillosis. This diagnostic imaging study demonstrated a more rapid clearance of pulmonary lesions in the rabbits treated with ABCD at 5 mg/kg/day than in rabbits treated with DAmB at 1 mg/kg/day (data not shown). The ABCD and DAmB regimens reduced the postmortem levels of *A. fumigatus* in tissues but did so at different rates. Thus, the differences in survival between rabbits treated with ABCD at 5 mg/kg/day and those treated with DAmB at 1 mg/kg/day were probably due to the combined effects of enhanced rate of tissue clearance, decreased pulmonary injury, and reduced nephrotoxicity in the ABCD group.

DAmB was more toxic and was associated with reduced survival. This diminished survival in rabbits receiving DAmB appeared to be related to nephrotoxicity and not to any comparative deficiency in antifungal activity. Although ABCD at 10 mg/kg/day produced the highest level of microbiological clearance and reduction of pulmonary lesions, it was also the most nephrotoxic dosage. Perhaps other modalities such as saline loading may improve renal perfusion and permit higher dosages of ABCD to be administered with reduced nephrotoxicity and greater antifungal activity (3, 16).

In summary, this study demonstrates that ABCD at 5 mg/kg/day achieved an optimal dosage with effective microbiological clearance, increased survival, and tolerable nephrotoxicity in a persistently granulocytopenic rabbit model of invasive pulmonary aspergillosis. These findings further add to the understanding of ABCD as it enters clinical trails for treatment of invasive pulmonary aspergillosis in granulocytopenic patients.

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