Micafungin (FK463) is a novel, semisynthetic antifungal echinocandin-like lipopeptide that inhibits the synthesis of 1,3β-glucan, an essential polymeric polysaccharide in the cell wall of many pathogenic fungi (12, 24). As a class, the echinocandins lack mechanism-based toxicity and have an extended spectrum of antifungal activity without cross-resistance to existing antifungal agents (4, 5, 9, 15, 18). In vitro, micafungin has demonstrated potent and broad-spectrum fungicidal activity against clinically relevant Candida spp. and potent inhibitory activity against Aspergillus spp. (21, 23, 25). The compound displayed promising antifungal efficacy in murine models of disseminated candidiasis as well as disseminated and pulmonary aspergillosis (16, 19, 20) and is currently in advanced stages of clinical development (12).

Little is still known, however, about the disposition of micafungin in plasma and tissues. Therefore, the purpose of this study was to assess the compartmental plasma pharmacokinetics and tissue distribution of micafungin at potentially therapeutic dosages in healthy laboratory animals. The information derived from this study will assist to further explore the relationships between concentration and effect of micafungin in pharmacodynamic models of disseminated candidiasis and invasive pulmonary aspergillosis in persistently neutropenic animals of the same species.


**MATERIALS AND METHODS**

**Experimental design.** (i) Study drug. Micafungin (FK463; Fujisawa USA, Inc., Deerfield, Ill.) was provided as a 10-mg/ml solution for injection and maintained at room temperature protected from light. Prior to use, the drug was freshly diluted with sterile normal saline to a 1-mg/ml solution. Micafungin was administered at ambient temperature as a slow intravenous (IV) bolus over 4 min through the indwelling catheter.

(ii) Animals. Healthy female New Zealand White rabbits (Hazleton, Denver, Pa.) weighing 2.8 to 3.2 kg were used in all experiments. They were individually housed and maintained with water and standard rabbit feed ad libitum according to the National Institutes of Health Guidelines for Laboratory Animal Care (2) and in fulfillment of American Association for Accreditation of Laboratory Animal Care criteria. Vascular access was established in each rabbit 72 h prior to experimentation by the surgical placement of a subcutaneous silastic central venous catheter as previously described (27).

(iii) Single-dose plasma pharmacokinetics. Three groups of three animals each were studied. Animals received micafungin at 0.5, 1, or 2 mg/kg of body weight as a steady IV bolus over 4 min. Plasma samples (2.0 ml of blood) were drawn immediately before administration of the drug and then at 0.1 (maximum...
concentration of drug in plasma. The concentration profiles (Cwa) at 0.25, 0.5, 1, 2, 4, 6, 8, 12, 18, and 24 h after the start of the IV bolus were determined using visual inspection of the plasma drug concentration-versus-time curves following single-dose administration of micafungin at 0.5 mg/kg. Accuracies in plasma were within 1.7 to 12.8%, and intra-assay variations were within 9.0% to 19.0% for nonplasma body fluids and tissue homogenates. The methods were sensitive to 0.05 mg/ml. The methods were sensitive to 0.05 mg/ml. Micafungin eluted at 10.3 to 24 h after dosing were not significantly different from those observed after administration of a single dose. Similar to single-dose administration, mean plasma drug levels fell below the LLQ (0.1 μg/ml) in a dose-dependent manner 8, 12, and 18 h after dosing. Consistent with dose-independent, linear plasma pharmacokinetics, total plasma clearance (CLt) and dose-normalized AUC0→∞ were not significantly different across the investigated dosage range. Similarly, the apparent volume of distribution at steady state (VSS) did not change with the dosage.

**RESULTS**

**Single-dose studies.** The estimated plasma drug concentration-versus-time curves following single-dose administration of micafungin are shown in Fig. 1A, and the corresponding mean compartmental pharmacokinetic parameters are listed in Table 1. IV bolus administration of micafungin at dosages of 0.5 to 2 mg/kg resulted in mean peak plasma drug levels that ranged from 7.67 ± 1.49 to 16.08 ± 1.72 μg/ml. Plasma drug concentration profiles showed a rapid initial distributive phase, followed by a slower elimination phase with an estimated elimination half-life of approximately 3 h. Mean plasma drug levels fell below the LLQ (0.1 μg/ml) in a dose-dependent manner 8, 12, and 18 h after dosing. Consistent with dose-independent, linear plasma pharmacokinetics, total plasma clearance (CLt) and dose-normalized AUC0→∞ were not significantly different across the investigated dosage range. Similarly, the apparent volume of distribution at steady state (VSS) did not change with the dosage.

**Multiple-dose studies.** The estimated plasma micafungin concentration-versus-time profiles following multiple daily doses of the compound for 7 days are shown in Fig. 1B, and the corresponding mean compartmental pharmacokinetic parameters are listed in Table 2. At all three dosage levels, plasma drug concentrations immediately prior to dosing were below the LLQ. Peak plasma drug concentrations immediately after dosing were not significantly different from those observed after administration of a single dose. Similar to single-dose administration, mean plasma drug levels fell below LLQ in a dose-dependent manner 8, 12, and 18 h postdosing. There were no significant differences in AUC (Fig. 2A), VSS, CLt, and half-life compared to the values after single doses. No differences in dose-normalized AUC0→∞ across the investigated dosages were noted by ANOVA and linear regression (Fig. 2B), indicating dose-independent plasma pharmacokinetics of micafungin also after multiple doses.

**Tissue distribution.** Mean tissue drug concentrations near peak plasma drug concentrations 30 min after the last of eight daily doses of micafungin are shown in Table 3. At this time point of the dosing interval, the highest concentrations were detected in the lung, followed by the liver, spleen, and kidney. Drug concentrations in these organs increased proportionally.

**Pharmacokinetic data analysis. (i) Pharmacokinetic modeling.** Pharmacokinetic parameters for micafungin were determined using compartmental analysis. Experimental plasma micafungin concentration-versus-time profiles were fitted to a two-compartment open model with IV bolus input and linear first-order elimination from the central compartment using iterative weighted nonlinear least-squares regression with the ADAPT II computer program (3). Model selection was guided by visual inspection of the plasma drug profiles and
administration of micafungin over 4 min. (A) Single-dose profiles after administration of 0.5, 1, and 2 mg/kg. (B) Profiles after administration of 0.5, 1, and 2 mg/kg over 7 days. Each point is the mean ± SEM for three rabbits at that time point. The LLQ was 0.100 μg/ml.

**FIG. 1.** Concentration-versus-time profiles in plasma after IV bolus administration of micafungin over 4 min. (A) Single-dose profiles after administration of 0.5, 1, and 2 mg/kg. (B) Profiles after administration of 0.5, 1, and 2 mg/kg over 7 days. Each point is the mean ± SEM for three rabbits at that time point. The LLQ was 0.100 μg/ml.

**TABLE 1. Single-dose compartmental pharmacokinetic parameters of micafungin in plasma**

<table>
<thead>
<tr>
<th>Drug dose (mg/kg)</th>
<th>C_{max} (μg/ml)</th>
<th>C_{min} (μg/ml)</th>
<th>AUC_{0→∞} (μg · h/ml)</th>
<th>V_{p} (liter/kg)</th>
<th>V_{c} (liter/kg)</th>
<th>V_{SS} (liter/kg)</th>
<th>CL_{p} (liter/h/kg)</th>
<th>CL_{c} (liter/h/kg)</th>
<th>α-HL (h)</th>
<th>β-HL (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.5</td>
<td>7.67 ± 1.495</td>
<td>&lt;LLQ</td>
<td>5.68 ± 0.43</td>
<td>0.30 ± 0.026</td>
<td>0.04 ± 0.008</td>
<td>0.301 ± 0.025</td>
<td>0.267 ± 0.047</td>
<td>0.09 ± 0.007</td>
<td>0.071 ± 0.003</td>
<td>2.97 ± 0.109</td>
</tr>
<tr>
<td>1.0</td>
<td>13.03 ± 0.204</td>
<td>&lt;LLQ</td>
<td>13.50 ± 1.56</td>
<td>0.251 ± 0.02</td>
<td>0.045 ± 0.000</td>
<td>0.296 ± 0.019</td>
<td>0.316 ± 0.008</td>
<td>0.077 ± 0.01</td>
<td>0.07 ± 0.002</td>
<td>3.2 ± 0.145</td>
</tr>
<tr>
<td>2.0</td>
<td>16.08 ± 1.725</td>
<td>&lt;LLQ</td>
<td>21.96 ± 1.73</td>
<td>0.254 ± 0.006</td>
<td>0.089 ± 0.018</td>
<td>0.343 ± 0.014</td>
<td>0.428 ± 0.023</td>
<td>0.089 ± 0.01</td>
<td>0.325 ± 0.02</td>
<td>3.04 ± 0.269</td>
</tr>
</tbody>
</table>

P value | 0.0113 | NA | 0.0005 | 0.9080 | 0.0007 | 0.2561 | 0.0255 | 0.5282 | 0.2566 | 0.7005 |

*a* All values represent the means ± SEMs for three rabbits. Abbreviations: V_{p} and V_{c}, volume of distribution in the peripheral and central compartment, respectively; CL_{p}, distributional clearance; α-HL, distributional half-life; β-HL, elimination half-life; NA, not applicable. The LLQ of the analytical assay was 0.1 μg/ml.

*b* P values for the comparison among dosage groups by ANOVA.
Vmean AUC0
dogs. In these species, after a single IV bolus of 1 mg/kg, the
single doses were similar to those obtained in mice, rats, and
ences in pharmacodynamics remains to be investigated.
dences in plasma pharmacokinetics are associated with differ-
for micafungin and caspofungin (14). Whether these differ-
multiple once-daily doses for 8 days revealed potentially therapeu-
therapies (14). Cognizant of the fact that different equilibria
may prevail during the dosing interval, assessment of tissue
concentrations of micafungin after the administration of mul-
tiple once-daily doses for 8 days revealed potentially therapeu-

0.086 ± 0.01 liter/h/kg, respectively, and were thus not different
from the values observed for micafungin following the identical
dosing schedule (11). In contrast, at similar dosages and dosing
schedules, anidulafungin exhibited an approximately sixfold-
lower mean Cmax, a twofold-faster CLt, a twofold-lower
AUC0–24, but a fourfold-larger VSS in comparison to the values
for micafungin and caspofungin (14). Whether these differ-
ences in plasma pharmacokinetics are associated with differ-
ences in pharmacodynamics remains to be investigated.
The plasma pharmacokinetics of micafungin in rabbits after
single doses were similar to those obtained in mice, rats, and
dogs. In these species, after a single IV bolus of 1 mg/kg, the
mean AUC0–24 ranged from 11.9 to 21.2 μg · h/ml, the mean
VSS ranged from 0.25 to 0.56 liter/kg, the mean CLt ranged
from 0.079 to 0.046 liter/h/kg, and the half-life ranged from
4.57 to 5.34 h, (S. Suzuki, M. Terakawa, F. Yokoyabashi, F.
volunteers, at dosages ranging from 12.5 to 50 mg given as a
2-h infusion by the IV route, micafungin exhibited linear phar-
macokinetics with mean Cmax ranging from 0.94 to 3.36 μg/ml
and mean AUC0–24 values of 17.11 to 60.93 μg · h/ml. The mean
VSS was between 0.237 and 0.242 liter/kg, and the terminal
half-life was approximately 15 h. While mean peak plasma
drug concentrations in humans were four- to 13-fold lower
than after bolus administration in rabbits, AUC0–24 values were
similar at the 12.5-mg dosage level (approximately 0.25 mg/kg)
and four- to fivefold higher at comparable dosages (J. Azuma,
I. Yamamoto, M. Ogura, T. Mukai, H. Suematsu, H.
F146, 1998). Notwithstanding the different modes of drug ad-
ministration, the plasma clearance of micafungin was approxi-
mately six- to sevenfold less in humans than that in rabbits. As
a consequence of the different modes of administration (i.e.,
bolus versus 2-h infusion), direct scaling of therapeutically
effective dosages from rabbits to humans would be best accom-
plished by formal pharmacodynamic models that link dosage,
concentrations over time, and antifungal effects.

Despite the fact that tissue drug concentrations represent a
mixture of drug concentrations in the intravascular, interstitial,
and intracellular compartments (1), information on these con-
centrations is of potential utility in the selection of antifungal
therapies (14). Cognizant of the fact that different equilibria
may prevail during the dosing interval, assessment of tissue
concentrations of micafungin after the administration of mul-
tiple once-daily doses for 8 days revealed potentially therapeu-

TABLE 2. Multiple-dose compartmental pharmacokinetic parameters of micafungin in plasma

<table>
<thead>
<tr>
<th>Drug dose (mg/kg)</th>
<th>Cmax (μg/ml)</th>
<th>Cmin (μg/ml)</th>
<th>AUC0–24 (liter/h/kg)</th>
<th>Vp (h)</th>
<th>Vc (liter/kg)</th>
<th>VSS (liter/kg)</th>
<th>CLt (liter/h/kg)</th>
<th>CLd (liter/h/kg)</th>
<th>α-HL (h)</th>
<th>β-HL (h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.5</td>
<td>9.44 ± 1.83</td>
<td>&lt; LLQ</td>
<td>6.22 ± 0.97</td>
<td>0.226 ± 0.053</td>
<td>0.031 ± 0.004</td>
<td>0.258 ± 0.057</td>
<td>0.182 ± 0.085</td>
<td>0.075 ± 0.012</td>
<td>0.07 ± 0.006</td>
<td>2.988 ± 0.424</td>
</tr>
<tr>
<td>1.0</td>
<td>18.09 ± 1.19</td>
<td>&lt; LLQ</td>
<td>14.16 ± 2.11</td>
<td>0.215 ± 0.02</td>
<td>0.029 ± 0.002</td>
<td>0.245 ± 0.02</td>
<td>0.235 ± 0.009</td>
<td>0.067 ± 0.01</td>
<td>0.063 ± 0.002</td>
<td>3.178 ± 0.192</td>
</tr>
<tr>
<td>2.0</td>
<td>19.16 ± 0.305</td>
<td>&lt; LLQ</td>
<td>22.29 ± 1.76</td>
<td>0.223 ± 0.011</td>
<td>0.090 ± 0.011</td>
<td>0.313 ± 0.007</td>
<td>0.393 ± 0.043</td>
<td>0.083 ± 0.013</td>
<td>0.101 ± 0.011</td>
<td>3.347 ± 0.447</td>
</tr>
</tbody>
</table>

P value* 0.0096 NA 0.0040 0.9774 0.0002 0.5479 0.0306 0.9227 0.0222 0.7995

* All values represent the means ± SEMs of 3 rabbits each. Abbreviations: Vp and Vc, volume of distribution of the peripheral and central compartment, respectively; CLt, distributional clearance; α-HL, distributional half-life; β-HL, elimination half-life; NA, not applicable. The LLQ of the analytical assay was 0.1 μg/ml.

A

B

FIG. 2. (A) AUC0–24 at the three investigated dosage levels (0.5, 1, and 2 mg) after administration of single and multiple doses of micafungin (FK). Each bar represents the mean ± SEM of three rabbits. Note the absence of drug accumulation in plasma over time after multiple once-daily doses. (B) Plot of dose-normalized AUC after multiple once-daily doses with micafungin over 7 days versus dosage. The values for individual animals (squares) and the corresponding means (triangles) ± SEM are shown. The slope of the regression line is not significantly different from zero, indicating linear disposition in plasma over the investigated dosage range.
tic drug concentrations in lung, liver, spleen, and kidneys near the completion of the initial distributive phase in plasma. Similar to amphotericin B (13), micafungin was undetectable in CSF and achieved relatively low levels in brain tissue compared to other sites. However, therapeutically effective levels of micafungin in brain tissue may be achieved in the state of tissue inflammation and/or necrosis, as evidenced by the effective clearance of Candida albicans from the central nervous system in our persistently neutropenic rabbit model of subacute disseminated candidiasis (V. Petraitis, R. Petraitiene, A. H. Groll, T. Sein, R. L. Schaufele, J. Bacher, and T. J. Walsh, Abstr. 40th Intersci. Conf. Antimicrob. Agents Chemother., abstr. 1684, 2000).

Micafungin achieved plasma and tissue drug concentrations that were severalfold in excess of the MICs at which 90% of the Candida and Aspergillus isolates tested are inhibited (21, 23, 25). In plasma, concentrations above these values were maintained in a dose-dependent manner for up to 18 h. Similar to cilofungin (26), caspofungin (7), and anidulafungin (10, 17, 22), micafungin exhibits predominantly concentration-dependent fungicidal activities against Candida spp. in vitro (Petraitis et al., 40th ICAAC). Concentration-dependent activity also was demonstrated in a Candida thigh infection model, where the ratio between tissue concentrations and MIC was found to be highly predictive for therapeutic efficacy of micafungin (S. Matsumoto, E. Warabe, Y. Wakai, Y. Koide, T. Ushitani, N. Teratani, K. Ohtomo, K. Hatano, F. Ikeda, T. Goto, F. Matsumoto, and S. Kuwahara, Abstr. 40th Intersci. Conf. Antimicrob. Agents Chemother., abstr. 1687, 2000). Pharmacokinetic and pharmacodynamic modeling of anidulafungin in our neutropenic rabbit model of disseminated candidiasis revealed that, apart from drug-specific threshold values for $C_{max}$, AUC$_{0-24}$, and tissue concentrations, maintenance of plasma drug concentrations above the minimum fungicidal concentration of the experimental isolate for $\geq 12$ h was associated with 100% efficacy (14). These findings and the documentation of a concentration-dependent, prolonged postantifungal effect of up to 12 h and longer for caspofungin and anidulafungin (6) suggest that once-daily dosing regimens are also appropriate for micafungin. Nevertheless, pharmacodynamic studies comparing single- versus split-dose regimens are needed for a pharmacodynamically founded determination of the optimal dosing regimen.

In conclusion, micafungin displayed linear plasma pharmacokinetics that were best described by a two-compartment pharmacokinetic model. The drug achieved and maintained potentially therapeutic plasma drug concentrations exceeding the MICs of susceptible opportunistic fungi and distributed into tissues that are common sites of deeply invasive infections. The compound was well tolerated without evidence of clinical or laboratory toxicity. The characterization of the pharmacokinetics of micafungin in the rabbit will be of help for the design of pharmacodynamic animal models investigating the concentration-response relationships of this novel echinocandin-like lipopeptide. The findings from such studies are anticipated to support the determination of optimal dosing regimens in patients.

**ACKNOWLEDGMENTS**

We thank Azhar Kalim at MDS Harris, Lincoln, Nebr., for assistance with the analytical assay of micafungin in plasma and our colleagues Myrna Candelario and Aida Field-Ridley for expert technical support in conducting these experiments.

**REFERENCES**


**TABLE 3. Tissue micafungin concentrations after multiple doses over 8 days**

<table>
<thead>
<tr>
<th>Drug dose (mg/kg)</th>
<th>Lung (µg/g)</th>
<th>Liver (µg/g)</th>
<th>Spleen (µg/g)</th>
<th>Kidney (µg/g)</th>
<th>Brain (µg/g)</th>
<th>CSF (µg/ml)</th>
<th>Choroid (µg/ml)</th>
<th>Vitreous humor (µg/ml)</th>
<th>Aqueous humor (µg/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.5</td>
<td>2.26 ± 0.10</td>
<td>2.05 ± 0.69</td>
<td>1.87 ± 0.09</td>
<td>1.40 ± 0.08</td>
<td>0.08 ± 0.01</td>
<td>ND</td>
<td>0.012 ± 0.014</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>1.0</td>
<td>5.59 ± 1.31</td>
<td>4.11 ± 0.15</td>
<td>4.30 ± 0.11</td>
<td>3.34 ± 0.14</td>
<td>0.10 ± 0.04</td>
<td>ND</td>
<td>0.027 ± 0.039</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>2.0</td>
<td>11.76 ± 1.40</td>
<td>8.82 ± 0.72</td>
<td>9.05 ± 0.25</td>
<td>6.12 ± 0.17</td>
<td>0.18 ± 0.02</td>
<td>ND</td>
<td>0.162 ± 0.096</td>
<td>ND</td>
<td>0.034 ± 0.032</td>
</tr>
</tbody>
</table>

* All values represent the means ± SEMs for three rabbits. Tissues and body fluids were obtained 30 min after the last of eight once-daily doses of FK463. ND, not detectable.