Intraocular Penetration of Intravenous
Micafungin in Inflamed Human Eyes

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Abstract

Eight eyes of 7 patients with fungal disease received intravenous injections of 150 to 300 mg micafungin, and samples of blood and cornea, retina-choroid, aqueous, and vitreous were collected. The micafungin levels in all collected samples exceeded the minimum inhibitory concentrations, however the levels in the vitreous and aqueous were lower. Our findings suggest that intravenous micafungin should be given in combination with intravitreal antifungal agents after vitrectomy in severe cases of intraocular fungal diseases.

Key words: micafungin; fungal endophthalmitis; vitrectomy; ocular penetration
Endogenous fungal endophthalmitis is a serious inflammatory disorder that commonly occurs in immunocompromised patients. Systemic treatments for endogenous fungal endophthalmitis include the polyenes and the triazoles (1,2). Micafungin is a water soluble echinocandin antifungal agent and it has comparable efficacy with fewer side effects as liposomal amphotericin B as a first-line treatment for candidaemia and invasive candidiasis (3). Micafungin can block the formation of biofilms of Candida spp (4), but has poor penetration into the vitreous after a single intravenous injection in animal models (5).

We investigated the intraocular penetration of intravenously administered micafungin in inflamed human eyes. Eight inflamed eyes of 7 patients (Table 1) that were scheduled to undergo vitrectomy or enucleation surgery were studied. The study protocol was approved by the Ethics Committees of Gifu University Hospital, and all patients signed an informed consent. Then, 150 mg or 300 mg/dose of micafungin was infused intravenously daily. Blood samples were collected approximately 1 hr after the last intravenous injection of micafungin, and the plasma was immediately separated from the whole blood by centrifugation. Ocular samples were obtained within 30 min (Case 1-5) or 90 min (Case 6 and 7) after collecting blood samples. All specimens were stored at -20º C until analyses. The aqueous and vitreous samples were not diluted.

The concentration of micafungin was measured by HPLC according to the methods described in detail by Yamato et al. and Niwa et al.(9, 10). Briefly, after the plasma proteins were precipitated with acetonitrile containing an internal standard, the analysis were separated on a TSK gel ODS-80 (150 mm × 4.6 mm internal diameter; Tosoh, Tokyo, Japan). Calibration curves were linear over the range of 0.01 to 2.5µg/mL for aqueous and vitreous, and of 0.1 to 25 µg/mL for plasma. The inter- and intra-day precision were below 15%, and the accuracy was within 15% at the quality controls.

Part of the vitreous and aqueous was cultured on Sabouraud agar, and the isolates were identified based on morphology and by sequencing the internal transcribed spacer region of the rDNA or the b-tubulin gene (11, 12). Candida albicans grew from the vitreous of Cases 1 and 2, Asperillus tubingensis from the vitreous in Cases 5, and Paecilomyces lilacinus in the corneal scraping of Case 6. Tests for drug susceptibility were performed on the vitreous isolates by broth microdilution according to the CLSI methods (13,14).

For the statistical analyses, unpaired t tests and Pearson product-moment correlation...
coefficients were used. A $P$ value of $<0.05$ was considered to be significant. All statistical analyses were performed using SPSS software version 16.0 (SPSS Japan, Tokyo, Japan.).

The concentrations of micafungin (mean±SD) was 21.02±4.59 µg/mL in the plasma, 0.10±0.07 µg/mL in the vitreous, and 0.08±0.12 µg/mL in the aqueous (Table 1). The mean concentrations for 150 and 300 mg micafungin were 17.02 and 23.30 µg/mL in the plasma ($P=0.035$; unpaired $t$ test), 0.05 and 0.10 µg/mL in the aqueous ($P=0.453$; unpaired $t$ test), and 0.09 and 0.10 µg/mL in the vitreous ($P=0.874$; unpaired $t$ test). There was no significant correlations between the total dose and the concentrations of micafungin in the plasma ($r=0.728$, $P=0.041$), in the aqueous humor ($r=0.400$, $P=0.373$), and in the vitreous humor ($r=0.513$, $P=0.194$).

The micafungin concentration in the cornea was 5.99 µg/g in Case 6 and 1.60 µg/g in Case 7. In Case 7, the drug concentration was 14.65 µg/g in the iris, 1.20 µg/g in the retina, and 5.81 µg/g in the choroid.

In 2 cases, the MICs of $C. albicans$ to amphotericin B, flucytosine, fluconazole, itraconazole, miconazole, voriconazole, and micafungin were 0.25 to 0.5, 0.25, 0.5 to 1, 0.125 to 0.25, 0.25, ≤0.015 to 8, and ≤0.03 µg/mL, respectively. In Case 5, the MICs of $A. tubingensis$ to amphotericin B, flucytosine, itraconazole, miconazole, voriconazole, and micafungin (Minimum Effective Concentration: MEC) were 1, 32, 1, 4, 1, and ≤0.015 µg/mL, respectively. In Case 6, the MICs of $P. lilacinus$ to amphotericin B, flucytosine, itraconazole, miconazole, voriconazole, and micafungin (MEC) were 2, >64, 0.5, 2, 0.125, and 0.25 µg/mL, respectively.

The guideline for the treatment of fungal endophthalmitis is systemic antifungal therapy combined with a monitoring of the endophthalmitis (15). In rabbits, Suzuki et al. reported that the concentration of micafugin in the retina-choroid and plasma exceeded the MICs for fungal pathogens after a single intravenous administration (5). The MIC$_{90}$ of micafungin for global surveillance was 0.06 µg/mL for $Candida$ spp., 0.06 µg/mL for $C. glabrata$ and $C. tropicalis$, and ≤0.008 µg/mL for $Aspergillus$ spp.(16). The MICs of micafungin against $C. albicans$ was ≤0.03 µg/mL and that for $A. tubingensis$ was ≤0.015 µg/mL (MEC) isolated in our hospital.

The fungal endophthalmitis may have led to the production of proinflammatory cytokines that could have disrupted the blood ocular barrier (17). However, the mean vitreous levels of micafungin with endogenous endophthalmitis still remained low suggesting that even after a disrupted blood-retinal barrier, micafungin penetrated poorly into the vitreous. Groll et al.
showed that the micafungin level in the aqueous of non-inflamed rabbit eyes was low after an intravenously injection (7). The concentration in the aqueous was not high even in our inflamed eyes except in an eye with severe anterior inflammation. The intraocular penetration may be related to differences in the molecular weights and the solubility in ocular fluids (18-20). The micafungin level in the iris was higher than that in other ocular tissues, suggesting that the drug in the anterior chamber was bound to the melanin in the iris.

The intravenous dosing regimen for invasive candidiasis with micafungin is 100 mg/day (15). The steady state plasma concentration of micafungin after repeated intravenously administrations is attained by 4 days, and the terminal half-life after the cessation of repetitive doses was 14 hour (21). We found a significant correlation between the total dose and the concentrations of micafungin in the plasma. However, the correlations between the concentration in the aqueous and vitreous humor after a single or total dose of micafungin were not significant. This may be due to differences in the sampling time and variations in the sampling procedures.

Our results showed that the concentration of micafugin in the cornea was high and exceeded the MICs for most Candida and Aspergillus spp (16). The micafugin in the cornea was from the tear film, aqueous, or limbal vessels. However, micafugin was not detected in the tear film in a case of tunnel fungal infection. Although the reason for the accumulation of micafugin in the cornea is unclear, the differences in the degree of protein binding or retention of the water-soluble echinocandin macromolecule in the proteoglycans of the corneal stroma may allow high levels of echinocandins to accumulate in the cornea (22).

In conclusion, our results showed that the concentrations of micafugin in the cornea, the iris, and the retina-choroid were higher than the MIC for Candida spp. and Aspergillus spp. We recommend that intravenous micafugin be considered only for patients with mild endogenous fungal endophthalmitis (isolated chorioretinitis without vitreous extension) without vitrectomy. It can also be considered as an alternative therapy for selected patients with C. glabrata endophthalmitis for whom first-line fluconazole therapy cannot be used because of drug resistance. Systemic administration of micafugin has good corneal penetration. Because the level of the micafugin in the aqueous and vitreous is not high, micafugin may be combined with an intravitreal antifungal agent with vitrectomy for the treatment of severe endogenous fungal endophthalmitis.
Author Disclosure Statement

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REFERENCES


TABLE1. Patient Demographics and Micafungin Concentrations in Plasma and Ocular Tissues

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Value or category for patient no.</th>
<th>Mean ± SD or ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Side</td>
<td>Left</td>
<td>Right</td>
</tr>
<tr>
<td>Sex</td>
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<td>Male</td>
</tr>
<tr>
<td>Age (years)</td>
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<td>67</td>
</tr>
<tr>
<td>Type of Surgery</td>
<td>PPV + PEA + IOL</td>
<td>PPV + PEA + IOL</td>
</tr>
<tr>
<td>Isolated Organism (sample)</td>
<td>Candida albicans (vireous)</td>
<td>Fungal endophthalmitis</td>
</tr>
<tr>
<td>Final Diagnosis</td>
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<td>Fungal endophthalmitis</td>
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<tr>
<td>Final BCVA (Before Surgery)</td>
<td>20/20 (12/20)</td>
<td>20/20 (6/20)</td>
</tr>
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<td>Others Medical Conditions</td>
<td>Myelodysplastic syndrome</td>
<td>Pyelonephritis</td>
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<tr>
<td>Micafungin</td>
<td>Dose (mg/day)</td>
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<tr>
<td>Total Dose (mg)</td>
<td>5700</td>
<td>3600</td>
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<tr>
<td>Duration (Days)</td>
<td>19</td>
<td>12</td>
</tr>
<tr>
<td>Plasma (µg/mL)</td>
<td>24.77</td>
<td>25.94</td>
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<tr>
<td>Aqueous (µg/mL)</td>
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<td>0.03</td>
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<td>Vitreous (µg/mL)</td>
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<td>Cornea (µg/g)</td>
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<tr>
<td>Iris (µg/g)</td>
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</tr>
<tr>
<td>Retina (µg/g)</td>
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<td>-</td>
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<tr>
<td>Choroid (µg/g)</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>


* Values in brackets show the each ocular tissue / plasma concentration ratios
* Reference no 20
* Poor vision has been detected 30 years ago
* After PPV+IOL removal
* Visual acuity before the vitrectomy and IOL removal
* Aphakic and avitreous eye
* 0.1% micafungin eye drop: for 9 days, 5 times per day