Mononuclear and Polymorphonuclear Leukocyte Dispositions of Clarithromycin and Azithromycin in AIDS Patients Requiring *Mycobacterium avium* Complex Prophylaxis

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The intracellular dispositions of clarithromycin and azithromycin in AIDS patients requiring *Mycobacterium avium* complex (MAC) prophylaxis were studied. The dispositions of both drugs in mononuclear and polymorphonuclear leukocytes were markedly different. Our data support the proven efficacy of these agents for MAC prophylaxis since clarithromycin and azithromycin displayed sustained intracellular concentrations which exceeded their MICs for MAC throughout the dosing periods.

*Mycobacterium avium* complex (MAC) is an opportunistic pathogen which resides in and replicates intracellularly in leukocytes, and as a result, disease due to this organism presents therapeutic challenges (8, 9, 15). However, owing to their extensive penetration of infected cells (5, 11, 14, 17), both clarithromycin (CLR) and azithromycin (AZM) have been found to be useful for the prevention of disease (7, 16). The concentrations of CLR and AZM in plasma are generally not maintained above their MICs for MAC; it is their intracellular concentrations at the site of infection that determine their clinical utility (2, 9, 14). The goal of this study was to characterize the leukocyte dispositions of CLR and AZM in AIDS patients requiring MAC prophylaxis (12).

This work was presented in part at the 36th Annual Meeting of the Infectious Diseases Society of America, Denver, Colo., 12 to 15 November 1998 [2a].

This was a prospective, open-labeled, two-way-crossover study. Patients were eligible for inclusion if they were >18 years of age, had no hypersensitivity to the study medications, were not infected with MAC, and were able to provide written informed consent. All procedures were reviewed and approved by the Institutional Review Board. Subjects were housed at the Clinical Research Center 12 h prior to commencement of blood sampling. Subjects fasted overnight and then were fed prior to medication administration. Subjects were administered either oral CLR (Abbott Laboratories, Abbott Park, Ill.) at 500 mg every 12 h or AZM (Pfizer Laboratories, New York, N.Y.) at 1,200 mg (two 600-mg tablets) every week. Each medication was started 4 weeks prior to sampling to attain steady-state concentrations. At the completion of the first sampling period, subjects were switched to the other medication with the normalization of all plasma data to 70 kg.

Plasma was used within 30 min of collection for the cell isolations. A volume of 100 μl of Red-Out (Robbins Scientific Corporation, Sunnyvale, Calif.) was added to each 10 ml of heparinized blood, and a gradient separation kit (1-Step Polybrene; Accurate Chemical & Scientific Corporation, Westbury, N.Y.) was used to simultaneously isolate the mononuclear (MN) and polymorphonuclear (PMN) cells.

The extraction and assay of CLR and AZM from plasma and cells were performed by previously validated high-performance liquid chromatography procedures (14, 17). The plasma AZM concentration range was 0.01 to 0.4 μg/ml, while the intrarun and interrun low-concentration (0.02 μg/ml) and high-concentration (0.3 μg/ml) quality control samples all had coefficients of variation (CVs) of <7%. The concentration range of intracellular AZM for the assay was 0.08 to 2 μg/ml, with intrarun and interrun CVs of <7%. A CLR concentration range of 0.1 to 4 μg/ml was used for both the plasma and intracellular determinations, and the intrarun and interrun low-concentration (0.2 μg/ml) and high-concentration (3 μg/ml) quality control samples had CVs of <4%. The OH metabolite of CLR was not assayed because it has only a marginal effect on the killing of MAC (2).

Because of the high purity of each sample and a lack of information on the differential uptake of these drugs into cellular subtypes, it was assumed that concentrations reflected the disposition of the predominant cells, not that of the subpopulations. The calculation used to more accurately determine the intracellular antibiotic concentration was modified from previous reports (6, 13). The pharmacokinetics of AZM and CLR in plasma and the intracellular compartment were calculated by a noncompartmental approach with the normalization of all plasma data to 70 kg.

Ten subjects (three females and seven males) were enrolled, and one male subject was withdrawn after he inadvertently took an extra dose of AZM. Overall, the adverse events related to the medication were minor; two subjects reported mild abdominal pain and cramping after taking AZM, and one subject reported an intermittent bitter metallic taste a week after starting CLR. The mean age (± standard deviation) was 43 ± 6.8 years, and the mean weight was 76.7 ± 30.2 kg. The median CD4+ cell count and RNA viral load were 179 cells/mm³ (range, 50 to 526) and 9,109 copies/mm³ (range, <400 to 189,774), respectively.

Table 1 shows the plasma and cellular pharmacokinetics of
AZM and CLR. The plasma AZM data from one subject were disregarded because of assay interference that could not be resolved by high-performance liquid chromatography manipulation. All other data are composites of the data for the remaining nine subjects. The half-life ($t_{1/2}$) of each medication in PMN cells was longer than that in MN cells, regardless of the regimen used. Figures 1 and 2 show the relationships of the concentrations in plasma and cells for each antibiotic. The high degree of tissue penetration resulted in concentrations in MN and PMN cells that were many times higher than those in plasma for both agents.

At present, limited data that characterize the steady-state intracellular dispositions of CLR and AZM in AIDS patients requiring MAC prophylaxis are available. The maximum concentration of drug in plasma ($C_{max}$) and the time to $C_{max}$ for CLR and AZM in this study were similar to data obtained with healthy volunteers; however, the observed $t_{1/2}$ and area under the concentration-time curve (AUC) were twice those obtained from studies performed with healthy volunteers (1, 3, 4). As a result of the apparently longer retention of these compounds in our patients, the $t_{1/2}$ is best regarded as an estimate since an extended sampling period would be required to accurately describe this parameter. However, since our patients required MAC prophylaxis, it was not justifiable to withhold therapy for the sole purpose of more accurately describing the pharmacokinetic profile. These data lend extra support to the observations that missed doses do not immediately lead to infection, since the $t_{1/2}$s in these reservoirs are prolonged. The ages and health statuses of our patients may have contributed to the altered pharmacokinetic values. Another likely cause for the plasma data discrepancy was the use of polypharmacy (data not shown) with antifungals, antiretrovirals, and protease inhibitors; drug interactions could magnify the interindividual variability.

This study confirms the finding of AZM enrichment in leukocytes from patients subjected to a once-a-week dosing regimen (6, 11, 18). Considering the pharmacokinetic profile and multiple dosing of AZM, it is not surprising to find that the AUC for AZM in MN and PMN cells is threefold higher than those measured for AIDS patients who received a single 1,200-mg dose (11). The mean intracellular/extracellular (I/E) AUC ratios were 8.7 and 13.1 for MN and PMN cells, respectively.

<table>
<thead>
<tr>
<th>Source and treatment</th>
<th>$k_{el}$ (h$^{-1}$)$^a$</th>
<th>$t_{1/2}$ (h)$^b$</th>
<th>$C_{max}$ (µg/ml)$^b$</th>
<th>$T_{max}$ (h)$^c$</th>
<th>AUC (mg · h/liter)$^d$</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0–12 h</td>
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<tr>
<td>Plasma</td>
<td></td>
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<tr>
<td>AZM$^d$</td>
<td>0.0107 ± 0.0040</td>
<td>77.2 ± 41</td>
<td>1.075 ± 1.390</td>
<td>1.5 (1–3.5)</td>
<td>24.1 ± 10.1</td>
</tr>
<tr>
<td>CLR</td>
<td>0.0855 ± 0.0349</td>
<td>9.27 ± 3.32</td>
<td>3.725 ± 1.78</td>
<td>4 (2–8)</td>
<td>38.3 ± 20.3</td>
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<td>MN cells</td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AZM</td>
<td>0.0096 ± 0.0054</td>
<td>106.3 ± 73.5</td>
<td>514 ± 322</td>
<td>2.5 (2.5–24)</td>
<td>41,592 ± 21,605</td>
</tr>
<tr>
<td>CLR</td>
<td>0.095 ± 0.061</td>
<td>10.1 ± 5.47</td>
<td>28.9 ± 11.2</td>
<td>4 (2–8)</td>
<td>297 ± 104</td>
</tr>
<tr>
<td>PMN cells</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>AZM</td>
<td>0.0046 ± 0.0033</td>
<td>168 ± 127.5</td>
<td>484 ± 194</td>
<td>8 (8–24)</td>
<td>65,358 ± 33,012</td>
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<tr>
<td>CLR</td>
<td>0.058 ± 0.037</td>
<td>20.1 ± 16.7</td>
<td>47.8 ± 29.1</td>
<td>2 (2–8)</td>
<td>470 ± 229</td>
</tr>
</tbody>
</table>

$^a k_{el}$, elimination rate constant; $T_{max}$, time to $C_{max}$.

$^b$ Means ± standard deviations.

$^c$ Means, with ranges in parentheses.

$^d$ Data for one subject excluded due to technical analysis problems.

**TABLE 1. Pharmacokinetics of AZM and CLR**

**FIG. 1. Pharmacokinetic profiles of CLR.** ■, MN cells; ○, PMN cells; ●, plasma.

**FIG. 2. Pharmacokinetic profiles of AZM.** ■, MN cells; ○, PMN cells; ●, plasma.
Interestingly, we found a similar relationship between our peripheral I/E ratio and those reported in bronchopulmonary-distribution studies of CLR in alveolar macrophages and epithelial lining fluid (5, 14, 17). Although the absolute concentration was higher within alveolar macrophages, our findings indicate that the intracellular disposition of CLR in both peripheral and pulmonary leukocytes is governed by the surrounding extracellular fluid concentrations. This information provides important insight into drug distribution characteristics and may allow further pharmacodynamic modeling of tissue sites based on peripheral plasma and cell data. While comparisons with AZM are difficult because it is generally not detected in the epithelial lining fluid, our I/E ratios were considerably different (300 to 2,700) than the I/E ratio of 40 reported by Rodvold et al. (17).

Our data support the proven efficacy of these agents and their respective dosing schedules for MAC prophylaxis since both displayed sustained intracellular concentrations exceeding their MICs for MAC for the entire dosing periods. In addition, these data support the concept that both agents provide a vast peripheral drug reservoir that may be sequestered at the site of infection.

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REFERENCES