ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Oct. 1996, p. 2375–2379 Vol. 40, No. 10
0066-4804/96/$04.00+0

Comparison of Bronchopulmonary Pharmacokinetics of Clarithromycin and Azithromycin

KALPANA B. PATEL,[1,2] DAWEI XUAN,[2] PAMELA R. TESSIER,[2] JOHN H. RUSSOMANNO,[3] RICHARD QUINTILIANI,[4] AND CHARLES H. NIGHTINGALE[4]*

Department of Pharmacy Practice, Arnold and Marie Schwartz College of Pharmacy, Long Island University, Brooklyn, New York 11201,[1] and Department of Pharmacy Research,[2] Lung Physicians of Central Connecticut, P.C.,[3] and Office of Research Administration,[4] Hartford Hospital, Hartford, Connecticut 06102-5037

Received 5 February 1996/Returned for modification 26 April 1996/Accepted 10 July 1996

The bronchopulmonary and plasma pharmacokinetics of clarithromycin (CLA; 500 mg given twice daily for nine doses) or azithromycin (AZ; 500 mg for the first dose and then 250 mg once daily for four doses) were assessed in 41 healthy nonsmokers. Bronchoalveolar lavage was performed at 4, 8, 12, or 24 h after administration of the last dose. The concentrations (mean ± standard deviation) of CLA, 14-hydroxyclarithromycin, and AZ were measured in plasma, epithelial lining fluid (ELF), and alveolar macrophage (AM) cells by high-performance liquid chromatography assay. The concentrations of CLA achieved in ELF were 34.02 ± 5.16 μg/ml at 4 h, 20.63 ± 4.49 μg/ml at 8 h, 23.01 ± 11.9 μg/ml at 12 h, and 4.17 ± 0.29 μg/ml at 24 h, whereas at the same time points AZ concentrations remained below the limit of assay sensitivity (0.01 μg/ml) for all but two subjects. The concentrations of CLA in the AM cells were significantly higher than those of AZ at 8 h (703 ± 235 and 388 ± 53 μg/ml, respectively). However, the ratio of the concentration in AM cells/concentration in plasma was significantly higher for AZ than for CLA for all time points because of the lower concentration of AZ in plasma. These results indicate that while AZ has higher tissue concentration to plasma ratios, as shown by other investigators, the absolute concentrations of CLA in AM cells and ELF are higher for up to 8 and 12 h, respectively, after administration of the last dose.

Clarithromycin (CLA), a new semisynthetic macrolide antibiotic, is indicated for the treatment of various infectious diseases and has gained its popularity in the treatment of respiratory tract infections owing to its high levels of microbiologic activity against *Streptococcus pneumoniae*, *Staphylococcus aureus*, *Haemophilus influenzae*, *Moraxella catarrhalis*, and other atypical organisms such as *Chlamydia pneumoniae*, *Legionella* species, and *Mycoplasma* species (5). In addition, the 14-hydroxy metabolite of CLA (14-OH CLA) is even more active than its parent compound against *H. influenzae* (9). CLA displays nonlinear pharmacokinetics and is widely distributed into human tissues and fluids (3, 7). A recent bronchopulmonary pharmacokinetic study of humans by Conte et al. (4) demonstrated that high concentrations of CLA and its metabolite were achieved in epithelial lining fluid (ELF) and alveolar macrophage (AM) cells.

Azithromycin (AZ), an azalide antimicrobial agent, is structurally different from the macrolides in that it has a 15-member rather than a 14-member ring. This structural modification allows for improved potency against gram-negative microorganisms, while it retains the efficacies of compounds with 14-member rings against gram-positive organisms. In addition, the pharmacokinetic profile of this agent is somewhat unique, with a longer half-life and greater distribution into tissues and fluids than the macrolides (6, 14). AZ readily penetrates phagocytic cells, allowing it to be useful in the treatment of infections caused by intracellular pathogens such as *Legionella pneumophila* and *Chlamydia trachomatis* (8, 12, 17, 21). Although the penetration ratios and concentrations in tissue of both AZ and CLA have been determined separately, a direct comparison between the two agents has never been published.

The purpose of the investigations described here was to measure and compare the concentrations of CLA, 14-OH CLA, and AZ in plasma, ELF, and AM cells in healthy volunteers at the end of a typical course of therapy for AZ and at steady-state levels for CLA.

MATERIALS AND METHODS

Study design. The present study was a randomized, prospective, nonblinded trial with healthy adult volunteers. All potential subjects were required to be 18 years old or older and within 10% of acceptable weight for their height according to the Metropolitan Life Insurance Company height/weight tables (16). Subjects were excluded from the study if any of the following conditions existed: a history of intolerance to macrolides, benzodiazepines, or lidocaine; the presence of clinically significant organ dysfunction; asthma; or a requirement to take chronic medications other than self-prescribed vitamins or birth control pills. Women were nonlactating and not pregnant.

After giving informed consent, 42 volunteers underwent a complete medical history, physical examination, and baseline laboratory testing, including but not limited to complete blood count; platelet count; and blood urea nitrogen, serum creatinine, aspartate aminotransferase, alanine aminotransferase, alkaline phosphatase, and total bilirubin concentration determinations. Female volunteers were given a serum pregnancy test. Twenty-one volunteers were randomized to both the CLA and AZ groups; the CLA group contained 12 males and 9 females; the AZ group contained 11 males and 10 females. The median ages (range) of the subjects in the CLA and AZ groups were 28 (21 to 41) and 29 (20 to 41) years, respectively. CLA (500-mg tablets) was administered orally every 12 h for nine doses. AZ (250-mg capsules) was administered orally at 500 mg on day 1 and then at 250 mg daily for four doses. Prior to the administration of the first dose a blood sample was collected. The first dose of the study medication was administered to subjects under direct supervision; this was followed by 30 min of observation for adverse events. Subsequent doses were taken according to verbal and written instructions and were documented in writing by the subjects. The subjects were also monitored via telephone to further assess any adverse events.

BAL. The subjects were randomized to undergo a standardized bronchoscopy and bronchoalveolar lavage (BAL) at either 4, 8, 12, or 24 h after the administration of the last dose of medication. The subjects were required to fast for at least 6 h before the bronchoscopy. The subjects were prepared with a 4% topical lidocaine spray to the posterior pharynx, and 2% lidocaine jelly was used in the nasal passageway for insertion of the bronchoscope. No systemic sedation was

* Corresponding author. Mailing address: Office of Research Administration, Hartford Hospital, 80 Seymour St., Hartford, CT 06102-5037. Phone: (203) 545-2865. Fax: (203) 545-5112.

used. A fiber-optic bronchoscope was inserted into the right middle lobe. A total of four 50-ml aliquots of normal saline were instilled into the right middle lobe, and each was immediately aspirated into a trap. The first aspirate was processed separately (first aspirate) from the second through the fourth aspirates, which were pooled (pooled aspirate). The volumes of the first and pooled aspirates were measured and recorded. Aliquots from the first and pooled aspirates were sent to the clinical laboratory for determination of the total leukocyte cell count and the percentage of AM cells of the total leukocyte cell count. The first and pooled aspirates were kept on ice until centrifugation at 400 × *g* for 5 min. The supernatants of both the first and pooled BAL fluid aspirates were immediately removed from the cell pellet, and all were frozen at −70°C until they were assayed for drug concentration(s). A small aliquot of both the first and the pooled BAL fluid supernatants were frozen at −70°C separately for urea concentration determination.

The blood pressure, pulse, respiratory rate, and heart rate of each subject were recorded prior to, at the completion of, and 30 to 60 min after the broncoscopy procedure. A blood sample for urea and drug concentration determination was also collected at the time of the bronchoscopy.

Specimen handling. Blood samples were spun at 1,000 × *g* for 10 min, and the plasma was separated and frozen until assay. An aliquot of plasma on the day of bronchoscopy was frozen separately for urea concentration determination. The cell pellets from the first and pooled BAL fluids were resuspended with a potassium phosphate buffer (pH 8.0) to a total volume of 10% of the recovered lavage fluid volume. The cell suspensions were freeze-thawed three times and were sonicated for 2 min before drug assay.

Drug assay. CLA, 14-OH CLA, and AZ were assayed by high-performance liquid chromatography (HPLC) techniques.

HPLC instrumentation. A Waters 510 pump (Waters Associates, Milford, Mass.) was equipped with a μ-Bondapak C_{18} column (3.9 by 150 mm; 10-μm particle size; Waters Associates). The analytes were detected with an ESA 5100A Coulochem electrochemical detector (Environmental Sciences Associates, Bedford, Mass.), which consisted of an ESA 5020 guard cell and an ESA 5010 dual-electrode analytical cell. The guard cell (+1.0 V) was placed in line before the injector to electrolyze components of the mobile phase and reduce the background current. The column effluent was monitored with the dual-electrode detector operated in the oxidative screen mode with applied potentials of +680 and +780 mV, respectively. Stainless steel in-line filters were placed before the guard cell and analytical cell to protect the porous electrodes. The samples were injected with a WISP-717 autosampler (Waters Associates). The detector signal was monitored with an HP3396 integrator.

HPLC mobile phase preparation. The mobile phase was a mixture of 0.05 M phosphate buffer (pH 7.5) and acetonitrile (45:55) filtered through a 0.22-μm-pore-size filter. The flow rate was 1.0 ml/min.

Sample extraction procedure. To a 200-μl aliquot of each sample, 50 μl of internal standard (for the AZ assay, CLA was used as the internal standard; for the CLA and 14-OH CLA assays, AZ was used as the internal standard) was added, and then each sample was alkalinized with 200 μl of 0.1 M sodium carbonate and vortexed for 30 s. The diluted sample was extracted with 3.5 ml of *tert*-methyl-butyl ether and was mixed for 20 min and then separated by centrifugation at 2,000 × *g* for 10 min. The ether layer was transferred to clean tubes and was evaporated to dryness at 37°C. The residue was reconstituted in 200 μl of the mobile phase and the components were mixed for 20 min; this was followed by centrifugation. The aqueous layer was transferred to injection vials, and 80-μl aliquots were injected into the HPLC system. Reagent-grade CLA and 14-OH CLA were provided by Abbott Laboratories (Abbott Park, Ill.). Reagent-grade AZ was provided by Pfizer Inc. (New York, N.Y.) Standard curves were prepared for the drugs in plasma, normal saline for the BAL fluid, and 10% plasma in 0.05 M sodium phosphate buffer (pH 8.0) for the AM cell suspension. The standard curves for CLA, 14-OH CLA, and AZ were constructed by weighted linear regression of the peak area ratio versus concentration and were linear in the following concentration ranges: for CLA, 0.1 to 4.0 μg/ml in plasma and 0.05 to 4.0 μg/ml in BAL fluid and AM cells; for 14-OH CLA, 0.2 to 5.0 μg/ml in plasma and 0.05 to 4.0 μg/ml in BAL fluid and AM cells; and for AZ, 0.01 to 2.0 μg/ml in plasma, 0.025 to 1.0 μg/ml in BAL fluid, and 0.01 to 1 μg/ml in AM cells. The lowest concentration of the prepared standards was used as the limit of quantification for each assay. If the concentration of a sample was above the range of the standard curve, a dilution of the sample was made in appropriate medium and the sample was reassayed. The final concentration was calculated by multiplying the concentration of the diluted sample obtained by the assay by the dilution factor. When a concentration of a sample was below the detection limit, it was not used in the statistical analyses.

The 14-OH CLA concentration in the BAL fluid was not measurable in any of the subjects because of the interference of lidocaine. There was no interference of lidocaine in the assays of 14-OH CLA, CLA, or AZ in AM cells or plasma or of CLA and AZ in BAL fluid.

To investigate the accuracy and precision of each assay, quality control samples (e.g., standards prepared identically to, but independently from, those of the standard curve) were assayed with each standard curve in each assay run. For all the drug assays, the concentrations obtained in all quality control samples were within 90 to 110% of expected values.

The variations between days (percent coefficient of variation) for AZ in BAL fluid for the quality control samples at 0.05 and 0.75 μg/ml were 4.1 and 7.4%, respectively; for the quality control samples at 0.05 and 0.5 μg/ml in AM cells they were 2.8 and 3.6%, respectively; and for the quality control samples at 0.2 and 1.5 μg/ml in plasma they were 3.0 and 6.5%, respectively. The coefficients of variation between days for the quality control samples with CLA at 0.5 and 3.0 μg/ml in BAL fluid were 4.2 and 3.6%, respectively. For the quality control samples with CLA at 0.5 and 3.0 μg/ml in AM cells, they were 4.5 and 4.1%, respectively; and for the quality control samples with CLA at 0.6 and 3.0 μg/ml in plasma, they were 6.1 and 5.0%, respectively. For the quality control samples at 0.5 and 3.0 μg/ml in AM cells, the coefficients of variation for 14-OH CLA were 6.3 and 2.7%, respectively, and for the quality control samples at 1.0 and 4.0 μg/ml in plasma they were 5.8 and 2.1%, respectively.

Estimation of ELF volumes and determination of drug concentrations in ELF and AM cells. The urea concentrations in the plasma samples were measured by the clinical laboratory at Hartford Hospital. The volume of ELF was determined by the urea dilution method (19). The urea concentration was measured in BAL fluid by a modified enzymatic assay (BUN kit UV-66; Sigma, Chemical Co., St. Louis, Mo.) (19). The volume of ELF in the BAL fluid was derived from the following relationships: $V_{ELF} = V_{BAL} \times (Urea_{BAL}/Urea_{Plasma})$, where V_{ELF} is the volume of ELF sampled by BAL, V_{BAL} is the volume of BAL fluid recovered, $Urea_{BAL}$ is the concentration of urea in BAL fluid, and $Urea_{Plasma}$ is the concentration of urea in the plasma. The concentration of antibiotic in the ELF $[ABX_{ELF}]$ was determined by the following relationship: $[ABX_{ELF}] = [ABX_{BAL}] \times (V_{BAL}/V_{ELF})$, where $[ABX_{BAL}]$ is the concentration of antibiotic in BAL fluid determined by HPLC.

The volume of AM cells in the cell pellet from the BAL fluid was determined from the absolute number of AM cells present. Total leukocyte cell counting was performed with a hemocytometer, and determination of the percentage of AM cells of the total leukocyte cells present was performed after centrifugation of the specimen in a cytocentrifuge (22). The volume of AM cells in the cell pellet suspension was calculated by using a mean macrophage cell volume of 2.42 $\mu l/10^6$ cells (2). The concentration of antibiotic in AM cells, $[ABX_{AM}]$ was calculated from the following relationship: $[ABX_{AM}] = ABX_{cell}/V_{AM}$, where ABX_{cell} is the amount of antibiotic in the cell pellet suspension determined by HPLC, and V_{AM} is the volume of AM cells in the cell pellet suspension (1).

Statistical analysis. Statistical evaluation of the effects of the site sampled (AM cells, the ratio of the concentration in AM cells/concentration in plasma, and plasma) and the sampling time (4, 8, 12, or 24 h) on the measured drug concentrations between the CLA and AZ groups was performed by an analysis of variance method with Sheffe's F test for multiple parameters. Likewise, the total leukocyte number, percent AM cells, and the concentrations of drug in the first and pooled aspirates were compared between the CLA and AZ groups. A P value of <0.05 was regarded as significant for all tests performed.

RESULTS

Forty-two subjects were enrolled in the study, and 41 subjects successfully completed the bronchoscopy procedure. Five subjects were placed into each of the CLA and AZ groups for each of the four sampling times with the exception of the CLA group sampled 12 h after drug administration, in which six subjects were enrolled. The one additional subject in the CLA group sampled 12 h after drug administration did not fast on the day of the bronchoscopy and was subsequently administered an additional nine doses of CLA (for a total of 18 doses). The data obtained for this subject were analyzed separately, but they were ultimately included in the final analysis since there was no difference between the results (z test; data not shown) for this subject and those for the other subjects. No serious adverse events were reported by the volunteers. One subject experienced transient bronchopulmonary spasms after the bronchoscopy procedure, but the spasms were relieved by the administration of albuterol via a metered dose inhaler. Another subject experienced transient chest discomfort after the bronchoscopy procedure, but the discomfort subsided over 24 h.

Both CLA and AZ concentrations in AM cells could be determined without interference from lidocaine in both the first and pooled aspirates from all subjects. Statistical analysis showed no significant differences between the concentrations of either drug in the first aspirate compared with those in the pooled aspirates. As a consequence, all aspirates were combined. The numbers (mean ± standard deviation) of AM cells recovered from the BAL aspirates (first and pooled aspirates combined) for the CLA and AZ groups were $5.4 \times 10^7 \pm 5.0 \times$

TABLE 1. CLA, 14-OH CLA, and AZ concentrations in plasma

Time of sample collection after last dose (h)	Concn in plasma (μg/ml [no. of subjects])[a]		
	CLA	14-OH CLA	AZ
4	3.29 ± 0.94 (5)[b]	1.43 ± 0.40 (5)[b]	0.09 ± 0.05 (5)
8	1.58 ± 0.50 (5)[b]	0.81 ± 0.25 (5)[b]	0.06 ± 0.05 (4)[c]
12	0.91 ± 0.59 (6)[b]	0.64 ± 0.37 (6)[b]	0.04 ± 0.02 (4)[c]
24	0.19 ± 0.09 (5)[b]	0.27 ± 0.04 (5)[b]	0.03 ± 0.03 (4)[c]

[a] Values are means ± standard deviations.
[b] Statistically significantly different from the concentration of AZ ($P < 0.05$).
[c] For one subject from each group, drug concentrations were below the limit of sensitivity of the assay (0.01 μg/ml); data for these subjects were not included in the calculation of the means.

10^7 (5.3×10^6 to 2.1×10^8) cells per liter and $3.2 \times 10^7 \pm 2.1 \times 10^7$ (1.9×10^6 to 8.2×10^7) cells per liter, respectively, which are recovery numbers that are not unusual for healthy subjects (20). The percentage (mean ± standard deviation) of AM cells among all leukocytes recovered from the BAL fluids (first and pooled combined) was 67.3% ± 14.4% for the CLA group and 66.7% ± 13.6% for the AZ group. No AMs were recovered in the pooled aspirate from one subject in the AZ group sampled 8 h after drug administration or in the first aspirate from one subject in the AZ group sampled 4 h after drug administration, and consequently, these data were not incorporated into the analysis of drug concentrations in AM cells. There were no statistically significant differences in the absolute number of AM cells recovered from the BAL fluid or the percentage of AM cells of total leukocyte cells in the BAL fluid between the two study groups.

The calculated volumes (mean ± standard deviation) of ELF in the BAL fluid from the first aspirates of all CLA and AZ subjects were 0.14 ± 0.09 and 0.17 ± 0.21 ml, respectively.

The calculated volumes (mean ± standard deviation) of ELF from the pooled BAL fluid aspirates of all samples from CLA and AZ subjects were 1.99 ± 1.27 and 2.15 ± 1.29 ml, respectively. No statistically significant differences in the volumes of ELF were observed.

The concentrations of CLA, 14-OH CLA, and AZ in plasma are presented in Table 1. At 4, 8, 12, and 24 h after the administration of the last dose, the concentrations of both CLA and 14-OH CLA were significantly higher than those of AZ. The concentrations of AZ in three subjects (one subject in each of the AZ groups sampled at 8, 12, and 24 h after drug administration) were below the limit of sensitivity of the assay.

The concentrations of CLA and AZ in the ELF samples are presented in Table 2. The CLA concentrations in the BAL fluid of several subjects were below the level of detection of the assay. The ratios of the mean concentration in ELF/mean concentration in plasma for CLA are also presented in Table 2. The concentrations of 14-OH CLA were not measurable because of the interference of lidocaine in the assay.

TABLE 2. Concentrations of CLA and AZ in ELF and ratio of concentration in ELF/concentration in plasma

Time of sample collection after last dose (h)[a]	CLA concn (μg/ml [no. of subjects])[b]	Ratio of concn in ELF/concn in plasma for CLA
4	34.02 ± 5.16 (5)	11
8	20.63 ± 4.49 (5)	14
12	23.01 ± 11.90 (5)	28
24	4.17 ± 0.29 (2)	31

[a] At the 4- and 24-h time points, AZ concentrations were below the limit of sensitivity of the assay (0.01 μg/ml) for all subjects. At the 8-h time point, AZ concentrations were below the limit of sensitivity of the assay (0.01 μg/ml) for four of five subjects, and for one subject the concentration in ELF was 1.93 μg/ml. At the 12-h time point, AZ concentrations were below the limit of sensitivity of the assay (0.01 μg/ml) for four of five subjects, and for one subject the AZ concentration in ELF was 1.76 μg/ml.
[b] Values are means ± standard deviations.

The concentrations of AZ in the BAL fluid recovered from subjects in all groups were notably low; only the pooled aspirates from two subjects (one in the group sampled at 8 h and one in the group sampled at 12 h after drug administration) had detectable levels. Therefore, calculation of the concentrations in ELF were not possible from the AZ-treated subjects.

Concentration-versus-time profiles for CLA, 14-OH CLA, and AZ in AM cells are presented in Table 3. The CLA concentrations in the AM cells were significantly higher than the AZ concentrations 8 h after administration of the last dose ($P < 0.05$). The 8-h time point for CLA concentrations includes data for the subject who received 18 doses of CLA instead of the nine doses required by the protocol. The concentrations were not different from those observed for the other subjects at the same time point; consequently, the data were included in the analysis. Also included in Table 3 is the ratio of the concentration in AM cells/concentration in plasma for CLA and AZ. The ratio for AZ was significantly higher than that for CLA at all time periods.

DISCUSSION

Recently, there has been increased interest in measuring the concentrations of antimicrobial agents at the site of the infection (1, 10, 15, 18). However, the in vivo determination of concentrations at these sites is plagued with certain methodological difficulties. First, calculation of intracellular drug concentrations must be done carefully. AMs must be separated quickly to minimize the efflux of drug from intracellular compartments to extracellular BAL fluid. Placing the samples on ice and centrifugation of the sample as quickly as possible help to diminish this loss. In addition, both CLA and AZ have slow rates of efflux from cells in general; thus, the loss to the BAL fluid would be expected to be minimal (21). Second, the accurate quantification of the number of macrophages is also important since high degrees of variability in the cell count will lead to variable concentrations (22). Most importantly, the determination of drug concentrations in the ELF is dependent on the calculation of the ELF volume from the recovered lavage fluid. The urea dilution method has been used with successful results, although the ELF volume may be overestimated, depending on the amount of time that the saline remains in the lungs (dwell time) (2, 19). Standardizing the BAL procedure will reduce this variability. Despite these methodological difficulties, the practice of measuring the drug concentrations at these sites continues to grow in popularity.

Only a few investigations of the pharmacokinetic properties of the newer macrolides in human pulmonary sites like AMs and ELF have been conducted and their results published (2, 4, 11). In addition, in vitro studies have indicated that the macrolides are not tightly bound to cellular components and readily diffuse into the extracellular space (12). Our investigation confirms the findings of Conte et al. (4) that the concentrations of CLA and 14-OH CLA are high and persist throughout the sampling period in both ELF and AM cells. CLA concentrations in ELF and AM cells are very high at the earlier time points and decrease over time, following the half-life of the drug in plasma, indicating that equilibrium exists between the extracellular and the intracellular compartments.

The AZ concentrations in plasma and ELF were less than

TABLE 3. CLA, 14-OH CLA, and AZ concentrations in AM cells and ratio of the concentration in AM cells/concentration in plasma

Time of sample collection after last dose (h)	Mean concn ± SD (μg/ml [no. of subjects])			Concn in AM cells/concn in plasma	
	CLA	14-OH CLA	AZ	CLA	AZ
4	1,996 ± 2,539 (5)	317 ± 310 (4)	450 ± 741 (5)	543[a]	1,292
8	703 ± 235 (5)[a]	256 ± 45 (2)	388 ± 53 (5)	465[a]	15,237
12	531 ± 299 (6)	124 ± 103 (5)	380 ± 573 (5)	1,041[a]	12,807
24	405 ± 299 (5)	117 ± 90 (4)	393 ± 364 (5)	1,265[a]	14,386

[a] Statistically significantly different from the concentration of AZ ($P < 0.05$).

half those of CLA. These results may be explained by the higher apparent volume of distribution of AZ, presumably caused by a large distribution of AZ to extracellular sites in comparison with that of CLA. Both CLA and AZ achieved high concentrations in the AM cells; however, the concentrations of AZ in the AM cells decreased slightly over the 24-h sampling period. This is probably due to the slow efflux of AZ from the cellular compartment into the extracellular fluid (13, 17).

Many investigators have used the penetration ratios of antimicrobial agents into various body sites and fluids to assess the clinical utilities of these agents. Indeed, as reported previously and confirmed by our results, AZ has a significantly higher ratio of concentration in AM cells/concentration in plasma than that of CLA, which is due largely in part to the low AZ concentrations observed in plasma in comparison with those of CLA. However, when one examines the actual concentrations in AM cells, both agents have high concentrations. Thus, the clinical utility of penetration ratios is somewhat misleading since bacterial eradication is a function of the drug concentration at the site of infection rather than penetration ratios.

Both CLA and AZ have successfully been used in the treatment of respiratory tract infections caused by typical bacterial pathogens like *S. pneumoniae*, *M. catarrhalis*, and *H. influenzae* and the atypical intracellular microorganisms including *Legionella* species, *C. pneumoniae*, and *Mycoplasma pneumoniae*. The excellent clinical responses of patients infected with intracellular pathogens to both agents is not surprising, because the intracellular concentrations of both drugs are exceedingly high in this compartment. In contrast, while CLA and its metabolite achieve effective concentrations in serum, the AZ concentrations in serum and ELF are below the typical MICs for *S. pneumoniae* and *H. influenzae*. Pharmacodynamic estimations of the area under the concentration-time curve/MIC, the time that the concentration is greater than the MIC, or maximum concentration of drug in serum/MIC, all predict unsatisfactory eradication rates; however, clinical data indicate that AZ is clinically effective against extracellular bacteria. This implies that the usual pharmacodynamic models for this drug may not fully explain the data, but the reasons for this observation, however, are still hypothetical. It might be due to the fact that the drug slowly diffuses out of the cell, resulting in high drug concentrations on the outer surface of the mammalian cell membrane, i.e., at the point of attachment of the pathogen to the solid surface of the cell, or it may involve the loading of granulocytes, which migrate to the local infection sites, go through an inflammatory burst, and dump their load of drug extracellularly, the so-called cellular drug delivery system. While these mechanisms are still speculative, it is doubtful that such mechanisms would apply only to AZ and not to CLA. Again, differences in drug behavior are speculated to be a function of differences in drug pharmacokinetics. The data indicate that there are pharmacokinetic differences between these drugs. While further study is required for a more complete understanding of this phenomenon, the data suggest that indexing the microbiological activities of these classes of agents to concentrations in plasma may not be an appropriate method of pharmacodynamic modeling except when the target organism's MIC is lower than the average drug concentration in serum, plasma, or interstitial fluid. In this case drug concentrations in the central compartment are so overwhelmingly high, compared with the MIC, that alternative models to explain the data are not needed. It should be mentioned, however, that the present study was performed with healthy volunteers, and the pharmacokinetic differences observed may be altered in patients with an inflammatory process in their lungs. As mentioned above, penetration ratios do not necessarily imply superior microbiological data in vitro or greater efficacy in vivo. Therefore, pharmacokinetic, microbiologic, and clinical data need to be integrated to derive a more complete pharmacodynamic model of the in vivo activities of macrolides and azelides if one is to rationally optimize therapeutic decision making.

ACKNOWLEDGMENTS

This work was supported by a grant from Abbott Laboratories.

We thank David P. Nicolau and the physicians at Lung Physicians of Central Connecticut, P.C., including Robert E. Mueller, R. Frederic Knauft, Michael M. Conway, and Eric T. Shore, as well as the staff of the Hartford Surgical Center, for invaluable assistance.

REFERENCES

1. **Baldwin, D. R., D. Honeybourne, and R. Wise.** 1992. Pulmonary disposition of antimicrobial agents: in vivo observations and clinical relevance. Antimicrob. Agents Chemother. **36:**1176–1180.
2. **Baldwin, D. R., R. Wise, J. M. Andrews, J. P. Ashby, and D. Honeybourne.** 1990. Azithromycin concentrations at the sites of pulmonary infection. Eur. Respir. J. **3:**888–890.
3. **Chu, S.-Y., D. S. Wilson, R. L. Deaton, A. V. Mackenthun, C. N. Eason, and J. H. Cavanaugh.** 1993. Single and multiple dose pharmacokinetics of clarithromycin, a new macrolide antimicrobial. J. Clin. Pharmacol. **33:**719–726.
4. **Conte, J. E., Jr., J. A. Golden, S. Duncan, E. McKenna, and E. Zurlinden.** 1995. Intrapulmonary pharmacokinetics of clarithromycin and of erythromycin. Antimicrob. Agents Chemother. **39:**334–338.
5. **Eliopoulous, G. M., E. Reiszner, M. J. Ferraro, and R. C. Moellering.** 1988. Comparative in vitro activity of A-56268 (TE-031), a new macrolide. J. Antimicrob. Chemother. **21:**671–675.
6. **Foulds, G., R. M. Shepard, and R. B. Johnson.** 1990. The pharmacokinetics of azithromycin in human serum and tissues. J. Antimicrob. Chemother. **25**(Suppl. A)**:**73–82.
7. **Fraschini, F., F. Scaglione, and G. Demartini.** 1993. Clarithromycin clinical pharmacokinetics. Clin. Pharmacokinet. **25:**189–204.
8. **Gladue, R. P., G. M. Bright, R. E. Isaacson, and M. F. Newborg.** 1989. In vitro and in vivo uptake of azithromycin (CP-62,993) by phagocytic cells: possible mechanisms of delivery and release at sites of infection. Antimicrob. Agents Chemother. **33:**277–282.
9. **Hardy, D. J., R. N. Swanson, R. A. Rode, K. Marsh, N. L. Shipkowitz, and J. J. Clement.** 1990. Enhancement of the in vitro and in vivo activities of clarithromycin against *Haemophilus influenzae* by 14-hydroxyclarithromycin, its major metabolite in humans. Antimicrob. Agents Chemother. **34:**1407–1413.

10. **Honeybourne, D., and D. R. Baldwin.** 1992. The site concentrations of antimicrobial agents in the lung. J. Antimicrob. Chemother. **30:**249–260.
11. **Honeybourne, D., F. Kees, J. M. Andrews, D. Baldwin, and R. Wise.** 1994. The levels of clarithromycin and its 14-hydroxy metabolite in the lung. Eur. Respir. J. **7:**1275–1280.
12. **Ishiguro, M., H. Koga, S. Kohno, T. Hayashi, K. Yamaguchi, et al.** 1989. Penetration of macrolides into human polymorphonuclear leukocytes. J. Antimicrob. Chemother. **24:**719–729.
13. **Johnson, J. D., W. L. Hand, J. B. Franis, N. King-Thompson, and R. W. Corwin.** 1980. Antibiotic uptake by alveolar macrophages. J. Lab. Clin. Med. **95:**429–439.
14. **Lalak, N. J., and D. L. Morris.** 1993. Azithromycin clinical pharmacokinetics. Clin. Pharmacokinet. **25:**370–374.
15. **Mandell, G. L.** 1994. Delivery of antibiotics by phagocytes. Clin. Infect. Dis. **19:**922–925.
16. **Metropolitan Life Insurance Company.** 1983. 1983 Metropolitan height and weight tables. Stat. Bull. **64:**2–9.
17. **Panteix, G., B. Guillaumond, R. Harf, A. Desbos, et al.** 1993. In-vitro concentration of azithromycin in human phagocytic cells. J. Antimicrob. Chemother. **31**(Suppl. E)**:**1–4.
18. **Prokesch, R. C., and W. L. Hand.** 1982. Antibiotic entry into human polymorphonuclear leukocytes. Antimicrob. Agents Chemother. **21:**373–380.
19. **Rennard, S. I., G. Basset, D. Lecossier, K. M. O'Donnell, P. Pinkston, P. G. Martin, and R. G. Crystal.** 1986. Estimation of volume of epithelial lining fluid recovered by lavage using urea as a marker of dilution. J. Appl. Physiol. **60:**532–538.
20. **Saltini, C., A. J. Hance, V. J. Ferrans, F. Basset, P. B. Bitterman, and R. G. Crystal.** 1984. Accurate quantification of cells recovered by bronchoalveolar lavage. Am. Rev. Respir. Dis. **130:**650–658.
21. **Steinberg, T. H.** 1994. Cellular transport of drugs. Clin. Infect. Dis. **19:**916–921.
22. **Wilcox, M., A. Kervitsky, L. C. Watters, and T. E. King, Jr.** 1988. Quantification of cells recovered by bronchoalveolar lavage. Am. Rev. Respir. Dis. **138:**74–80.