Comparative pharmacodynamics of moxifloxacin and levofloxacin in an in vitro dynamic model: prediction of the equivalent AUC/MIC breakpoints and equiefficient doses


*Department of Pharmacokinetics, Centre for Science and Technology LekBioTech, 8 Nauchny proezd, Moscow 117246, Russia; †Department of Medicine, Mount Auburn Hospital, Cambridge, MA, USA

To demonstrate the impact of the different pharmacokinetics of moxifloxacin and levofloxacin on their antimicrobial effects (AMEs), killing and regrowth kinetics of two clinical isolates of Staphylococcus aureus and one each of Escherichia coli and Klebsiella pneumoniae were studied. With each organism, a series of monoeponential pharmacokinetic profiles of single doses of moxifloxacin (T1/2 = 12.1 h) and levofloxacin (T1/2 = 6.8 h) were simulated. The respective eight-fold ranges of the ratios of area under the concentration–time curve (AUC) to the MIC were 58–475 and 114–934. Species- and strain-independent linear relationships observed between the intensity of AME (IE) and log AUC/MIC were not superimposed for moxifloxacin and levofloxacin (R² = 0.99 in both cases). The predicted AUC/MIC ratios for moxifloxacin and levofloxacin that might be equivalent to Schentag’s AUC/MIC breakpoint for ciprofloxacin (125) were estimated at 80 and 130, respectively. The respective equivalent MIC breakpoints were 0.41 mg/L (for a 400 mg dose of moxifloxacin) and 0.35 mg/L (for a 500 mg dose of levofloxacin). Based on the IE–log AUC/MIC relationships, equiefficient 24 h doses (D24s) of moxifloxacin and levofloxacin were calculated for hypothetical strains of S. aureus, E. coli and K. pneumoniae with MICs equal to the respective MIC50s (weighted geometric means of reported values). To provide an ‘acceptable’ IE = 200 (log cfu/mL)•h, the D24s of moxifloxacin for all three organisms were much lower (150, 30 and 60 mg, respectively) than the clinically proposed 400 mg dose. Although the usual dose of levofloxacin (500 mg) would be in excess for E. coli and K. pneumoniae (D24 = 36 and 220 mg, respectively), it might be insufficient for S. aureus (the estimated D24 = 850 mg). Moreover, to provide the same effect as a 400 mg D24 of moxifloxacin against staphylococci, levofloxacin would have to be given in a 5000 mg D24, which is 10-fold higher than its clinically accepted dose. The described method of generalization of data obtained with specific organisms to other representatives of the same species might be useful to predict the AMEs of new quinolones.

Introduction

Moxifloxacin (BAY 12-8039) has been studied extensively using in vitro dynamic models but only four studies were comparative. At least three studies exposed differentially susceptible organisms to a given clinical dose and, therefore, to different ratios of the area under the concentration–time curve to the MIC (AUC/MIC). However, the resulting AUC/MIC ranges were only partly overlapping (moxifloxacin versus grepafloxacin, sparflaxon and levofloxacin) or did not overlap at all (moxifloxacin versus ciprofloxacin). Moreover, in most cases the simulated AUC/MICs of moxifloxacin were shifted towards higher values than those of the comparators and, therefore, moxifloxacin quite expectedly showed greater effects. Despite these limitations, these data still allow comparison of the quinolones in terms of AUC/MIC–response relationships, but the AUC/MIC analysis of the effect was not performed or did not show reasonable relationships. In three non-comparative studies with moxifloxacin more...
or less distinct AUC/MIC relationships with the antimicrobial effect were established, but the equiefficient AUC/MIC ratio and equiefficient dose could not be predicted because of the lack of a comparator.

Such predictions may be based on an analysis of bacterial strain- and species-independent AUC/MIC relationships with the intensity of the antimicrobial effect (IE, the area between control growth and bacterial killing/regrowth curves) and the species-specific dose–response relationships as established over a wide range of AUC/MICs. In the present study, experiments that include the complete regrowth phase were used to compare the antimicrobial effects of moxifloxacin and levofloxacin on Staphylococcus aureus, Escherichia coli and Klebsiella pneumoniae in an in vitro dynamic model.

Materials and methods

Antimicrobial agents and bacterial strains

Moxifloxacin and levofloxacin (kindly provided by Bayer Corporation, West Haven, CT, USA and Ortho-McNeill Pharmaceuticals, Raritan, NJ, USA, respectively) were used in the study.

Two clinical isolates of S. aureus with different susceptibilities to moxifloxacin and levofloxacin and one each of E. coli and K. pneumoniae were selected for the study. The true MICs for these organisms determined by multiple serial dilutions as described elsewhere were 0.18 mg/L (S. aureus 944), 0.37 mg/L (S. aureus 916), 0.10 mg/L (E. coli 11557) and 0.32 mg/L (K. pneumoniae 56) of moxifloxacin and 0.25, 0.60, 0.20 and 0.20 mg/L, respectively, of levofloxacin. For the prediction of the antimicrobial effects of these quinolones on hypothetical representatives of the above-mentioned species, weighted geometric means of the reported MIC₅₀s of moxifloxacin and levofloxacin were calculated. The respective geometric values of the MIC₅₀ of moxifloxacin for S. aureus, E. coli and K. pneumoniae were 0.15, 0.03 and 0.06 mg/L, and those of levofloxacin were 0.7, 0.03 and 0.18 mg/L, respectively.

In vitro dynamic model and simulated pharmacokinetic profiles

A dynamic model described previously was used in the study. The operation procedures, reliability of simulations of the quinolone pharmacokinetic profiles and the high reproducibility of the time–kill curves provided by the model have been reported elsewhere.

A series of monoexponential profiles that mimic single dose administration of moxifloxacin and levofloxacin were simulated. The simulated half-lives (12.1 h for moxifloxacin and 6.8 h for levofloxacin) represented weighted means of the values reported in humans: 9.1–13.4 h and 6.0–7.4 h, respectively. The respective rates of fresh nutrient medium influx into the 60 mL (moxifloxacin) or 40 mL (levofloxacin) central compartments and the antibiotic- and bacteria-containing medium efflux from this compartment were 3.4 and 4.1 mL/h, respectively.

The mean simulated AUC/MIC ratios of moxifloxacin and levofloxacin were similar to those used in our previous studies with trovafloxacin and ciprofloxacin. In the present study, experiments that include the complete regrowth phase were used to compare the antimicrobial effects of moxifloxacin and levofloxacin on Staphylococcus aureus, Escherichia coli and Klebsiella pneumoniae in an in vitro dynamic model.

Determination of the time–kill curves and the antimicrobial effect

In each experiment multiple sampling of medium containing bacteria from the central compartment was performed throughout the observation period. The duration of the experiments was defined in each case as the time until antibiotic-exposed bacteria reached the maximum numbers observed in the absence of antibiotic (≥10⁶ cfu/mL). The lower limit of the viable counts was 2 × 10³ cfu/mL. The procedure used for quantification of viable counts has been reported elsewhere.

As described earlier, the antimicrobial effect (E) at each time point (t) was expressed by the difference between logarithms of the respective viable counts in the control growth curve (Nₓ) and in the time–kill curve (Nₐ): E(t) = log Nₓ – log Nₐ (Figure 2). Either the area between the log Nₓ–t and log Nₐ–t curves (Figure 2a) or the area under the E–t curve (Figure 2b) describes the total antimicrobial effect.
Moxifloxacin versus levofloxacin pharmacodynamics

Effect as expressed by \( I_E \). The upper limit of bacterial numbers, i.e., the cutoff level on the regrowth and control growth curves used to determine the \( I_E \) was \( 10^{11} \text{ cfu/mL} \). In case of lower counts, they were extrapolated to the cutoff level by using a logistic function.

Relationships between the effect and the AUC/MIC or dose

The \( I_E \) versus log AUC/MIC data sets obtained with each quinolone against \( S. aureus, E. coli \) and \( K. pneumoniae \) were fitted by the equation \( I_E = a + b \log \text{AUC/MIC} \) (Equation 1).

When predicting the AUC/MIC breakpoints for moxifloxacin and levofloxacin, the reported breakpoint value for ciprofloxacin, 125, which correlated with bacterial eradication in patients with respiratory tract infections, was used. This reference breakpoint reflects the critical value of the area under the inhibitory curve (AUIC), which is similar to the AUC/MIC.

To express the antimicrobial effects as a function of quinolone dose (\( D \)), the AUC in the linear relationship between \( I_E \) and log AUC that corresponds to Equation 1 written for a given quinolone–pathogen pair was substituted by \( D \) according to the linear equation: \( \text{AUC} = c D \) (Equation 2). The values of \( c \) for moxifloxacin and levofloxacin (0.08 and 0.11, respectively) were calculated on the basis of reported pharmacokinetic data that were obtained with moxifloxacin (\( Ds \) from 50 to 800 mg) and levofloxacin (\( Ds \) from 100 to 1000 mg).

Correlation and regression analyses of the relationship between \( I_E \) and log AUC/MIC for each quinolone were performed at a level of significance of \( P = 0.05 \).

Results

The time courses of viable counts that reflect killing and regrowth of \( S. aureus, E. coli \) and \( K. pneumoniae \) exposed to monoexponentially decreasing concentrations of moxifloxacin and levofloxacin as well as the respective control growth curves are shown in Figure 3a–h. As seen in Figure 3, the time–kill curves observed with both quinolones against these three bacterial species yielded similar patterns. At the AUC/MIC ratios studied, regrowth followed a rapid and considerable reduction in bacterial numbers. Steep ascending branches of the \( E–t \) curves (Figure 3i–p) reflect the rapid onset of the antimicrobial effect. The maximal \( E_s (E_{max}s) \) produced by both quinolones were greater at higher AUC/MICs, although the AUC/MIC-induced differences of \( E_{max}s \) were less pronounced than those of the time shift of the regrowth phase (Figure 3a–h) and of the descending branches of the \( E–t \) curves (Figure 3i–p). As seen in Figure 3, these shifts were distinctly dependent on the simulated AUC/MIC: the higher the AUC/MIC, the later the regrowth or the disappearance of the antimicrobial effect.

The respective \( I_Es \) correlated well with log AUC/MICs for both moxifloxacin and levofloxacin (Figure 4). The \( I_E–\log \text{AUC/MIC} \) plots fitted by Equation 1 were linear, bacterial species and strain independent but quinolone specific (\( r^2 > 0.99 \) in both cases). The effects produced by the same AUC/MIC ratio of moxifloxacin were greater than those of levofloxacin. For example, at AUC/MIC = 250, the \( I_E \) of moxifloxacin was 45% higher than that of levofloxacin.

Based on the \( I_E–\log \text{AUC/MIC} \) (Equation 1) and AUC–\( D \) (Equation 2) relationships, the respective dose–response curves were constructed for each quinolone against hypothetical representatives of \( S. aureus, E. coli \) and \( K. pneumoniae \) with MICs equal to the geometric means of MIC50s (Figure 5). The same antimicrobial effect [\( I_E = 200 \) (log cfu/mL)] against \( S. aureus \) might be provided by a much lower absolute dose of moxifloxacin than of levofloxacin: 150 versus 850 mg, respectively (Figure 5c). On the other hand, the \( D_{24} \) of levofloxacin that would reach the same
A. A. Firsov et al.

effect as produced by a 400 mg $D_{24}$ of moxifloxacin, would be 10-fold higher (5000 mg) than the usual clinical dose of levofloxacin. Based on the extrapolated relationship between $D$ and AUC of levofloxacin, the 5000 mg $D_{24}$ is out of the actual $D$ range in the AUC–$D$ set fitted by Equation 2, and this latter estimate is more conditional than the other values shown in Figure 5. However, it does reflect the order of difference between the quinolone doses that might be necessary to provide the same antimicrobial effect. The doses of moxifloxacin and levofloxacin necessary to efficiently suppress the growth of *E. coli* (30 and 36 mg, respectively) and *K. pneumoniae* (60 and 220 mg, respectively) are much lower than 400 mg of moxifloxacin or 500 mg of levofloxacin.

**Discussion**

This comparative pharmacodynamic study provides dose–response relationships and prediction of equiefficient doses of moxifloxacin and levofloxacin for *E. coli, K. pneumoniae* and *S. aureus*. These predictions are based on bacterial strain- and species-independent AUC/MIC relationships of the total antimicrobial effect ($I_E$) for the novel quinolone (moxifloxacin) and the comparator (levofloxacin) ($r^2 = 0.99$ in both cases).

Previously reported AUC/MIC relationships using alternative integral endpoints of moxifloxacin’s effect were either weaker ($r^2 0.51–0.69$ with a linear model$^8$ and $r^2$ 0.78–0.92 with a sigmoidal model$^1,4$) or uncertain.$^7$ This

---

**Figure 3.** The kinetics of killing and regrowth of bacteria exposed to moxifloxacin (a–d, open symbols) and levofloxacin (e–h, filled symbols) and the respective *E–t* curves (i–l and m–p). Bacteria were *S. aureus* 944 (a, e, i, m), *S. aureus* 916 (b, f, j, n), *E. coli* 11557 (c, g, k, o) and *Klebsiella pneumoniae* 56 (d, h, l, p). The extrapolated portions of the time–kill curves are indicated by dotted lines. The number on each curve indicates the simulated AUC/MIC ratio.
Moxifloxacin versus levofloxacin pharmacodynamics

might have resulted in part from intrinsic limitations inherent in the endpoints, area under bacterial time–kill curve (AUBC)\textsuperscript{1,4,8} and area above the time–kill curve (AAC)\textsuperscript{7} as discussed elsewhere.\textsuperscript{31} For example, unlike I\textsubscript{E}, AUBC may underestimate the antimicrobial effect at small AUC/MICs and overestimate it at large AUC/MICs when regrowth may not be seen within the observation period. It is not by chance that the correlations reported with AUBC calculated from zero time to 48 h (AUBC\textsubscript{0–48}) appeared to be stronger ($r^2$ 0.92) than those with AUBC calculated from zero time to 24 h (AUBC\textsubscript{0–24}, $r^2$ 0.78)\textsuperscript{4} because the former endpoint more completely considered the regrowth phase than the latter. Such an analysis is not applicable to the data reported in the other studies cited\textsuperscript{1,8} where regrowth of four of six strains of \textit{S. aureus} and \beta-haemolytic streptococci\textsuperscript{8} and eight of nine strains of \textit{Haemophilus influenzae} and \	extit{Moraxella catarrhalis}\textsuperscript{4} did not occur during the 48 h observation period.

The usefulness of long-term observations that include the entire regrowth phase rather than only initial bacterial

\begin{figure}
\centering
\includegraphics[width=\textwidth]{figure4}
\caption{(a) AUC/MIC-dependent antimicrobial effects of moxifloxacin (open symbols, bold line) and levofloxacin (filled symbols, thin line) on \textit{S. aureus} 944 (●, ▲), \textit{S. aureus} 916 (○, ▼), \textit{E. coli} 11557 (□, ■) and \textit{K. pneumoniae} 56 (◇, ●) fitted by Equation 1: $a = -415$ and $b = 323$ (moxifloxacin), $a = -178$ and $b = 178$ (levofloxacin). Italicized numbers indicate the $I_E$s corresponding to an AUC/MIC of 250. (b) Species-independent $I_E$–log AUC/MIC plots for moxifloxacin and levofloxacin based on data from this study and ciprofloxacin (dashed line) based on reported data.\textsuperscript{10} The equivalent AUC/MIC breakpoints are indicated by italicized numbers.}
\end{figure}

\begin{figure}
\centering
\includegraphics[width=\textwidth]{figure5}
\caption{Dose-dependent antimicrobial effects of moxifloxacin (bold lines) and levofloxacin (thin lines) on hypothetical strains of (a) \textit{E. coli} (MIC\textsubscript{50} of moxifloxacin and levofloxacin 0.03 mg/L), (b) \textit{K. pneumoniae} (MIC\textsubscript{50} of moxifloxacin and levofloxacin 0.06 and 0.18 mg/L, respectively) and (c) \textit{S. aureus} (MIC\textsubscript{50} of moxifloxacin and levofloxacin 0.15 and 0.7 mg/L, respectively). Italicized numbers indicate the doses that provide the same $I_E$s.}
\end{figure}
killing in establishing AUC/MIC–response relationships has been demonstrated with trovafloxacin and ciprofloxacin.10,11,31 In this light, the lack of reported correlations between the AUC/MIC ratio of levofloxacin and ciprofloxacin and the time to a 1000-fold reduction in starting inoculum (T99.9%) in experiments with Streptococcus pneumoniae41 and between doses of several quinolones and T99.9% with S. aureus42 might be expected. Similarly, minimal, if any, dose-induced changes could be seen in the rate and extent of initial killing of S. pneumoniae and Enterococcus faecalis exposed to moxifloxacin.24 In another study, although initial killing of K. pneumoniae was similar with moxifloxacin and trovafloxacin, the patterns of bacterial regrowth were quite different.25 Thus, the establishment of AUC/MIC–response relationships may depend dramatically on the experimental design and the method of quantification of the antimicrobial effect.

The I_E–log AUC/MIC relationships established here for moxifloxacin and levofloxacin revealed greater antimicrobial effects with moxifloxacin at a given AUC/MIC ratio. Similar differences were reported in our studies with trovafloxacin and ciprofloxacin.10,11 By comparing the I_E–log AUC/MIC relationships for moxifloxacin and levofloxacin with that for ciprofloxacin,10 AUC/MIC breakpoints can be predicted that might be equivalent to Schentag’s AUC/MIC = 125 established in a clinical setting.38 As seen in Figure 4b, to provide an ‘acceptable’ I_E = 200 (log cfu/mL)-h that corresponds to the AUC/MIC = 125 for ciprofloxacin, the equivalent AUC/MIC breakpoint for moxifloxacin might be lower, at 80, and that for levofloxacin might be higher, at 130, than the breakpoint value for ciprofloxacin. As follows from Equation 2, a single dose of moxifloxacin (400 mg) provides its AUC of 33 mg•h/L. Its MIC breakpoint is equal to 33/80 = 0.41 mg/L. The respective value for a 500 mg dose of levofloxacin is lower: 46.5/130 = 0.35 mg/L. These MIC breakpoints are higher than the geometric means of MIC95% of moxifloxacin and levofloxacin for E. coli (0.03 mg/L for both agents) and K. pneumoniae (0.06 and 0.18 mg/L, respectively). Most strains of S. aureus will not be covered by moxifloxacin (geometric mean of MIC95% 0.7 mg/L) whereas they are more likely to be covered by moxifloxacin (geometric mean of MIC95% 0.15 mg/L).

Although the relevance of these estimates may be verified only in a clinical setting, together with the equiefficient doses, they might be useful for in vitro comparison of relative efficacies of the quinolones. In particular, these data support the important role of the longer half-lives of the newer extended spectrum quinolones whose pharmacokinetic profiles result in a greater antimicrobial effect.10,11

Acknowledgement

This study was supported by a grant from the Bayer Corporation.

References


Moxifloxacin versus levofloxacin pharmacodynamics


Received 28 January 2000; returned 2 May 2000; revised 1 June 2000; accepted 5 August 2000