ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Jan. 1995, p. 170–174 Vol. 39, No. 1
0066-4804/95/$04.00+0

Pharmacokinetics and Tolerance of DU-6859a, a New Fluoroquinolone, after Single and Multiple Oral Doses in Healthy Volunteers

MITSUYOSHI NAKASHIMA,[1] TOSHIHIKO UEMATSU,[1] KAZUHIRO KOSUGE,[1] KAZUO UMEMURA,[1] HIDEO HAKUSUI,[2] AND MAKOTO TANAKA[2]*

Department of Pharmacology, Hamamatsu University School of Medicine, Hamamatsu, Shizuoka 431-31,[1] and Drug Metabolism and Analytical Chemistry Research Center, Developmental Research Laboratories, Daiichi Pharmaceutical Co. Ltd, 1-16-13 Kitakasai, Edogawa-ku, Tokyo 134,[2] Japan

Received 28 March 1994/Returned for modification 12 July 1994/Accepted 21 October 1994

The pharmacokinetics and tolerance of DU-6859a, 7-[(*7S*)-7-amino-5-azaspiro[2,4]heptan-5-yl]-8-chloro-6-fluoro-1-[(*1R, 2S*)-2-fluoro-1-cyclopropyl]-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid sesquihydrate, were investigated in healthy male Japanese volunteers after single (25, 50, 100, and 200 mg) and multiple (100 mg three times a day for 6 days plus once a day on the 7th day and 50 mg every 12 h for 13 doses) oral doses. DU-6859a was well tolerated at all doses, and all 36 subjects completed the study; mild transient soft stool in five volunteers and mild transient diarrhea in one volunteer on the multiple-dose (100 mg three times a day) study were the only side effects reported. No drug crystals were observed in the urine after the single 200-mg dose and the 100-mg three times a day regimen. DU-6859a was rapidly absorbed in the fasted state. The mean maximum concentration in serum (C_{max}) ranged from 0.29 to 1.86 μg/ml for the 25- to 200-mg dose, and the mean time to reach C_{max} ranged from 1.0 to 1.3 h. The terminal half-life ranged from 4.4 to 5.0 h. The area under the curve increased dose dependently. The serum protein binding of the drug was approximately 50%. The apparent volume of distribution clearly exceeded 1 liter/kg, suggesting good tissue penetration. Within 48 h, the cumulative urinary recovery of unchanged drug amounted to 69 to 74% of the dose administered, while fecal excretion up to 48 h after the 200-mg dose accounted for ca. 3% of the dose. Food intake did not affect the rate and extent of absorption of DU-6859a to a clinically significant extent. During multiple oral dosing, the accumulation of the drug in serum was close to the theoretically predicted values, which indicated that there was virtually no drug accumulation.

7-[(*7S*)-7-Amino-5-azaspiro[2,4]heptan-5-yl]-8-chloro-6-fluoro-1-[(*1R, 2S*)-2-fluoro-1-cyclopropyl]-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid sesquihydrate (DU-6859a) is a new fluoroquinolone antimicrobial agent with a broad spectrum and high in vitro activity against various aerobic and anaerobic gram-positive and gram-negative organisms (3). Its chemical structure is shown in Fig. 1. DU-6859a is currently under phase 2 clinical investigation in Japan. Early pharmacokinetics and tissue distribution studies in different animal species revealed that this new fluoroquinolone was rapidly and well absorbed following oral administration and that drug levels in most tissues except the central nervous system exceeded the corresponding levels in serum (2). In rats, DU-6859a was extensively conjugated with glucuronic acid to form its ester glucuronide (1a). On the other hand, the serum radioactivity concentrations after a single oral administration of [^{14}C]DU-6859a to dogs and monkeys were close to serum levels of unchanged drug determined by the high-pressure liquid chromatography (HPLC) method, which indicated that DU-6859a underwent metabolism to a very limited extent in these animal species (5).

In this study, the pharmacokinetics and tolerance of DU-6859a after single and multiple oral doses in healthy Japanese male volunteers have been investigated.

(Part of this research was presented at the 33rd Interscience Conference on Antimicrobial Agents and Chemotherapy, New Orleans, La., 17 to 20 October 1993.)

* Corresponding author.

MATERIALS AND METHODS

Test compound. Daiichi Pharmaceutical Co. Ltd. (Tokyo, Japan) supplied 25- and 50-mg tablets of DU-6859a. DU-6859 represents the anhydrate of DU-6859a (Fig. 1). The doses were expressed as the equivalents of DU-6859.

Subjects. Thirty-six healthy male Japanese volunteers participated in the study after being fully informed of the purpose and risks involved. They gave written informed consent to participate in the study, which was approved by the Ethical Committee of Hamamatsu University School of Medicine. The subjects ranged in age from 24 to 48 years and weighed between 52.8 and 87.0 kg. They were judged to be healthy, based on a physical examination and standard biochemical, hematological, and urinalysis screening tests. All subjects were free of other medications for at least 1 week before and during the study. Alcohol and caffeine-containing beverages were prohibited during the study.

Dosing and sample collection. For the single-dose study, 24 subjects were assigned to one of four groups of six. After overnight fasting (unless otherwise stated), groups 1 to 4 received a 25-, 50-, 100-, or 200-mg dose of DU-6859, respectively. The drug was administered orally with 100 ml of water at about 0900 h. Lunch was provided after 4 h, and an evening meal was provided after 9 h. The subjects were allowed to drink water freely. In the study where the effect of food on the pharmacokinetics of DU-6859a was to be investigated, group 3, which was given the 100-mg dose, received the same dose 30 min after a standard breakfast. The standard breakfast consisted of two rolls, 8 g of margarine, 25 g of cheese, a medium-boiled egg, 100 ml of orange juice, and 150 ml of low-fat milk. Group 3 ingested the drug in the nonfasting state 10 days later. In addition to the blank samples taken shortly before drug administration, serial blood samples (5 ml) were collected 0.5, 1, 1.5, 2, 3, 4, 6, 8, 12, 24, 36, and 48 h after oral dosing. The blood samples were allowed to stand at room temperature for 30 min and were centrifuged at 2,000 × *g* for 15 min to separate serum. Urine samples were collected from 0 to 2, 2 to 4, 4 to 6, 6 to 8, 8 to 12, 12 to 24, and 24 to 48 h. Fecal samples were collected for 2 days in the single-200-mg-dose study. All samples were stored frozen at −20°C until analysis.

For the multiple-dose study, the remaining 12 subjects were assigned to two groups of six. Group 5 received DU-6859a at a dose of 100 mg three times a day (t.i.d.) at 0900, 1500, and 2100 h for 18 doses (days 1 to 6) and once on day 7. The other group (group 6) received DU-6859a at a dose of 50 mg every 12 h (twice a day [b.i.d.] at 0900 and 2100 h) for 13 doses (days 1 to 7). For group 5, serial blood samples (5 ml) were collected predosing and 0.5, 1, 2, 3, 4, 6, 8, 12, 24, 48,

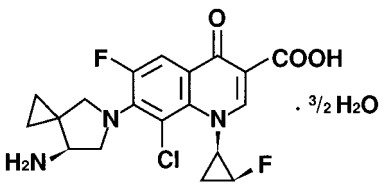

FIG. 1. Chemical structure of DU-6859a.

72, 73, 74, 75, 76, 78, 80, 84, 96, 120, 144, 145, 146, 147, 148, 150, 152, 156, 168, 192, and 216 h after the initial oral dose. Serum samples were obtained in the same way as in the single-dose study. Urine samples were collected from 0 to 24, 24 to 48, 48 to 60, 60 to 72, 72 to 74, 74 to 76, 76 to 78, 78 to 80, 80 to 82, 82 to 96, 96 to 120, 120 to 144, 144 to 156, 156 to 168, 168 to 180, 180 to 192, and 192 to 216 h. For group 6, serial blood samples (5 ml) were collected predosing and 0.5, 1, 1.5, 2, 3, 4, 6, 8, 12, 24, 48, 72, 73, 74, 75, 76, 78, 80, 84, 96, 120, 144, 145, 146, 147, 148, 150, 152, 156, 168, and 192 h after the initial dose. Urine samples were collected at 0 to 2, 2 to 4, 4 to 6, 6 to 8, 8 to 12, 12 to 24, 24 to 48, 48 to 60, 60 to 72, 72 to 96, 96 to 120, 120 to 144, 144 to 156, 156 to 168, and 168 to 192 h. All samples were stored at −20°C until analysis. Microscopic urinalysis to detect the presence of drug crystals was performed on fresh urine samples taken predosing and 2, 4, and 6 h after the single 200-mg dose and 2, 4, and 6 h after the 10th dose (day 4) during repeated oral doses (100 mg t.i.d.).

Analytical method. The concentrations of DU-6859 in serum and urine were determined by using an HPLC method (1). In brief, DU-6859 and the internal standard were extracted from serum and urine by means of a Bond Elut C8 LRC column. The extracts were chromatographed on a reversed-phase column using tetrahydrofuran–50 mM KH_2PO_4 (adjusted to pH 2 by adding phosphoric acid)–1 M ammonium acetate (19:81:1, vol/vol) as the mobile phase at a flow rate of 1.0 ml/min. DU-6859 eluted from the analytical column was irradiated with UV light in an on-line photochemical reactor. Fluorescence detection at an excitation wavelength of 280 nm and at an emission wavelength of 430 nm resulted in a detection limit of 0.01 μg/ml for serum and 0.1 μg/ml for urine. The intraday and interday accuracy and precision, as indicated by relative error and coefficient of variation, respectively, were within 15%.

Fecal specimens were homogenized with 50 mM KH_2PO_4 (500 or 1,000 ml, depending on weight). The homogenate was centrifuged at 1,800 × *g* for 15 min, and the supernatant was then processed through the same analytical procedures as used for the serum and urine samples. There were no interfering peaks on the chromatograms of the fecal extracts. The calibration curve was linear within the range from 0.092 to 23.5 μg/ml of supernatant.

All drug concentrations were expressed as equivalents of DU-6859 (anhydrate).

Serum protein binding. The binding of DU-6859 to serum proteins was determined by the ultrafiltration method (MPS-3; Amicon Corp.). DU-6859 did not adsorb to the membrane. The binding study was conducted on serum samples obtained from the volunteers at three time points (1, 4, and 8 h after dosing) in the 100-mg dose study. The unbound fraction (FU) in serum was calculated as the concentration in the filtrate divided by the original serum concentration. The serum protein binding rate was calculated as (1 − FU) × 100.

Pharmacokinetic and statistical analysis. Visual inspection of the log-linear concentration-time curves revealed that the serum concentrations of DU-6859 declined biphasically. The drug concentration-time data for each volunteer were fitted individually to a two-compartment open model with first-order absorption with or without lag time, corresponding to the equation $C(t) = Ae^{-\alpha(t - t_0)} + Be^{-\beta(t - t_0)} - Ce^{-K_a(t - t_0)}$, where $C(t)$ (in micrograms/milliliter) is the estimated concentration of DU-6859 at time t; A and B (in micrograms/milliliter) are the zero time intercept of the distribution (α) phase and elimination (β) phase, respectively, and α and β (per hour) represent the slopes of the α and β phases, respectively; C is equal to $A + B$; K_a (per hour) is the absorption rate; and t_0 is the lag time. Calculations were performed with the nonlinear least-squares computer program MULTI developed by Yamaoka et al. (7). No weighting factor was used. The goodness of fit of the model with or without lag time was determined by visual inspection of the difference between the observed values and the simulated curve. The terminal elimination half-life was calculated as 0.693/β. Maximum serum concentration (C_{max}) and the time to reach C_{max} (T_{max}) were obtained from the measured values. The area under the concentration-time curve (AUC) was determined to the last quantifiable serum concentration using the linear trapezoidal rule and extrapolated to infinity using the terminal-phase rate constant. The mean residence time was calculated as the ratio of the area under the first-moment curve (AUMC) to the AUC_{0-24}. The apparent total body clearance (Cl_{tot}/F) was calculated by using the equation Cl_{tot}/F = dose/$AUC_{0-\infty}$. The renal clearance (Cl_R) was calculated as $[X]/AUC_{0-24}$, where $[X]$ is cumulative urinary excretion of DU-6859 from 0 to 24 h after oral dosing. The simulation of the concentration-time profiles of DU-6859 after multiple oral dosing was performed based on the pharmacokinetic parameters calculated for the single 100-mg oral dose in the non-fasted state.

For statistical comparison between the pharmacokinetic parameters obtained in the fasted state and those obtained in the non-fasted state, a paired Student's *t* test and Wilcoxon signed rank test were used, with $P = 0.05$ considered the minimal level of significance. The Wilcoxon signed rank test was used only for T_{max}. To evaluate the correlation between the doses ranging from 25 to 200 mg and the resulting AUC and C_{max}, linear regression analysis was performed. All data are expressed as mean ± standard deviation (SD) ($n = 6$).

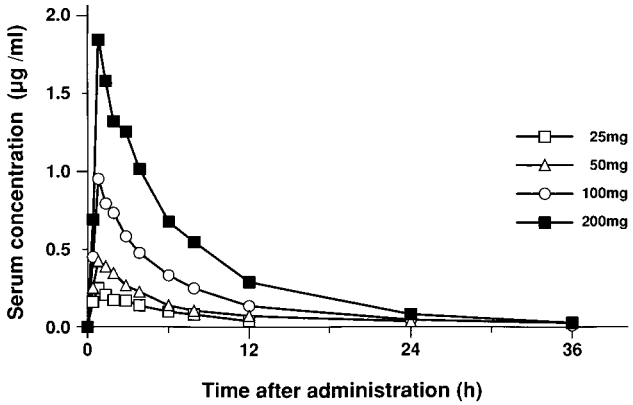

FIG. 2. Mean serum concentration-time profiles for DU-6859 in six fasted healthy male volunteers after single oral doses of 25, 50, 100, and 200 mg.

RESULTS

Safety and tolerance. All 36 volunteers completed the single- and multiple-dose studies. Mild transient soft stool in five volunteers and mild transient diarrhea in one volunteer in the group receiving multiple oral 100-mg doses were the only side effects observed. No drug crystals were detected in the urine after the single 200-mg dose and multiple doses (100 mg t.i.d.). None of the adverse events were considered serious. There were no clinically remarkable changes in serum chemistry, hematology, or urinalysis test results during the study.

Single-dose pharmacokinetics. The mean drug concentrations in serum versus time profiles obtained after single oral administration of DU-6859a (25, 50, 100, and 200 mg) to fasted Japanese male volunteers are shown in Fig. 2. The kinetic profiles could be fitted to a two-compartment open model with first-order absorption with or without lag time. The pharmacokinetic parameters for each dose are summarized in Table 1. The lag times ranged from 0.23 to 0.58 h for all doses studied. The absorption of DU-6859a from the empty gastrointestinal tract was rapid, and C_{max} values of 0.29, 0.51, 1.00, and 1.86 μg/ml of serum appeared within 1 to 2 h after administration. After C_{max} was reached, the serum drug level decreased biphasically, with elimination half-lives of 4 to 5 h. The half-lives proved to be independent of the dose. The volume of distribution at steady state (V_{ss}/F) ranged from 1.46 to 1.88 liters/kg.

The serum protein binding rates 1, 4, and 8 h after a single oral dose of 100 mg were (mean ± SD) 54.5% ± 3.0%, 47.8% ± 6.1%, and 46.0% ± 6.8%, respectively. The $AUC_{0-\infty}$ and C_{max} increased dose dependently. A good linear correlation was found between the administered doses and the resulting AUC and C_{max}, with correlation coefficients of 0.929 and 0.952, respectively.

The cumulative urinary excretion of unchanged drug (collection period, 0 to 48 h) amounted to 69.2% ± 5.9%, 73.6% ± 6.8%, 73.3% ± 11.9%, and 72.4% ± 4.6% of the dose after a single oral administration of 25, 50, 100, and 200 mg, respectively. Cl_R ranged from 185 to 240 ml/min and remained almost constant as the dose increased. The fecal excretion of unchanged drug after a single oral 200-mg dose was investi-

TABLE 1. Pharmacokinetic parameters of DU-6859a after a single oral dose to healthy male volunteers

Parameter	Mean ± SD (n = 6)			
	25-mg dose	50-mg dose	100-mg dose	200-mg dose
C_{max} (μg/ml)	0.29 ± 0.08	0.51 ± 0.14	1.00 ± 0.14	1.86 ± 0.36
T_{max} (h)	1.3 ± 0.9	1.2 ± 0.5	1.2 ± 0.5	1.0 ± 0.0
K_a (h^{-1})	4.13 ± 2.01	6.12 ± 5.99	7.76 ± 8.37	5.09 ± 4.89
Lag time (h)	0.58 ± 0.64	0.39 ± 0.35	0.31 ± 0.24	0.23 ± 0.25
$t_{1/2}$ (h)	4.40 ± 0.35	4.62 ± 1.29	5.02 ± 1.96	4.60 ± 0.80
V_{ss}/F (liters/kg)	1.46 ± 0.26	1.88 ± 0.46	1.84 ± 0.37	1.79 ± 0.25
$AUC_{0-\infty}$ (μg · h/ml)	1.52 ± 0.32	2.62 ± 0.53	5.55 ± 1.22	12.03 ± 3.27
MRT (0–24 h) (h)	5.87 ± 1.15	5.93 ± 0.51	5.83 ± 0.45	6.18 ± 0.68
Cl_{tot} (ml/min)	284 ± 54	328 ± 59	313 ± 72	293 ± 76
Cl_R (ml/min)	185 ± 19	240 ± 31	227 ± 57	211 ± 65

gated, and 3.10% ± 0.48% of the dose given was recovered up to 48 h postdosing.

To investigate the effect of food on the pharmacokinetics of DU-6859a, six volunteers (group 3) received an oral 100-mg dose in the nonfasted state. The serum concentration-time profiles are shown in Fig. 3. Food intake had no significant influence ($P > 0.05$) on C_{max}, AUC, T_{max}, V_{ss}/F, elimination half-life, or Cl_R or Cl_{tot} of DU-6859. There was a significant increase in MRT in nonfasted subjects (6.46 ± 0.44 h) compared with that in fasted subjects (5.83 ± 0.45 h).

The mean urinary concentration-time profiles of DU-6859 up to 48 h after oral dosing are shown in Fig. 4. The urinary concentrations increased dose dependently. The concentrations in the sample obtained 12 to 24 h after oral doses of 25, 50, 100, and 200 mg were 5.3 ± 1.0, 9.0 ± 1.8, 14.1 ± 4.1, and 41.8 ± 11.8 μg/ml, respectively.

Multiple-dose pharmacokinetics. A plot of the mean observed and predicted serum concentration-time profile data for each dosage is shown in Fig. 5. The simulation was performed by using the pharmacokinetic parameters obtained for a single 100-mg oral dose to nonfasted subjects. Good agreement was found between the observed and predicted values over the 7-day dosing period, and visual inspection of the pharmacokinetic profiles showed that the steady-state concentrations of DU-6859 were achieved on the second day in both dosage groups. In group 5 (100 mg t.i.d. for 19 doses), the mean peak concentrations after the 1st, 10th, and 19th doses were 0.67 ± 0.12, 0.82 ± 0.18, and 0.89 ± 0.11 μg/ml, respectively, which were achieved 2 h after each dosing. The mean trough drug levels in serum ranged from 0.26 to 0.32 μg/ml and did not change over the 7 days. In group 6 (50 mg every 12 h for 13 doses), the mean peak concentrations after the 1st, 7th, and 13th doses were 0.35 ± 0.05, 0.49 ± 0.04, and 0.45 ± 0.07 μg/ml, respectively, with T_{max} being reached 2 h after each dose. The mean trough drug levels in serum ranged from 0.07 to 0.12 μg/ml and were almost constant over the 7 days.

The mean urinary excretion of unchanged drug during mul-

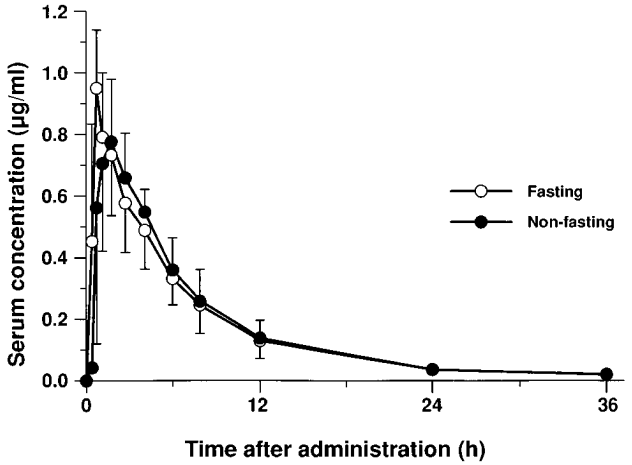

FIG. 3. Serum concentration-time profiles for DU-6859 in six fasted and nonfasted healthy male volunteers after a single oral dose of 100 mg. Error bars indicate SD (n = 6).

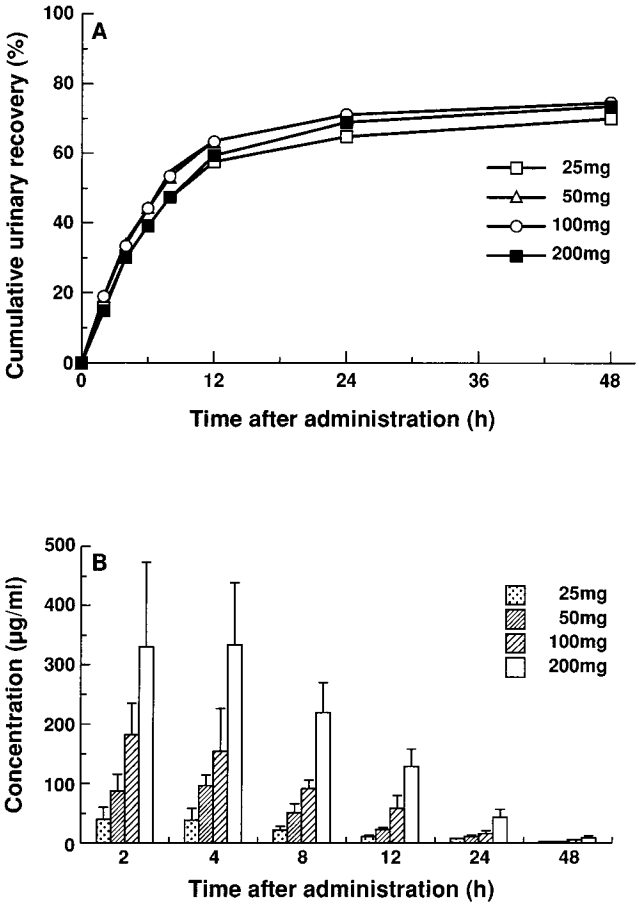

FIG. 4. (A) Urinary concentrations and (B) cumulative urinary excretion of DU-6859 in six fasted subjects after single oral doses of 25, 50, 100, and 200 mg. Error bars indicate SD (n = 6).

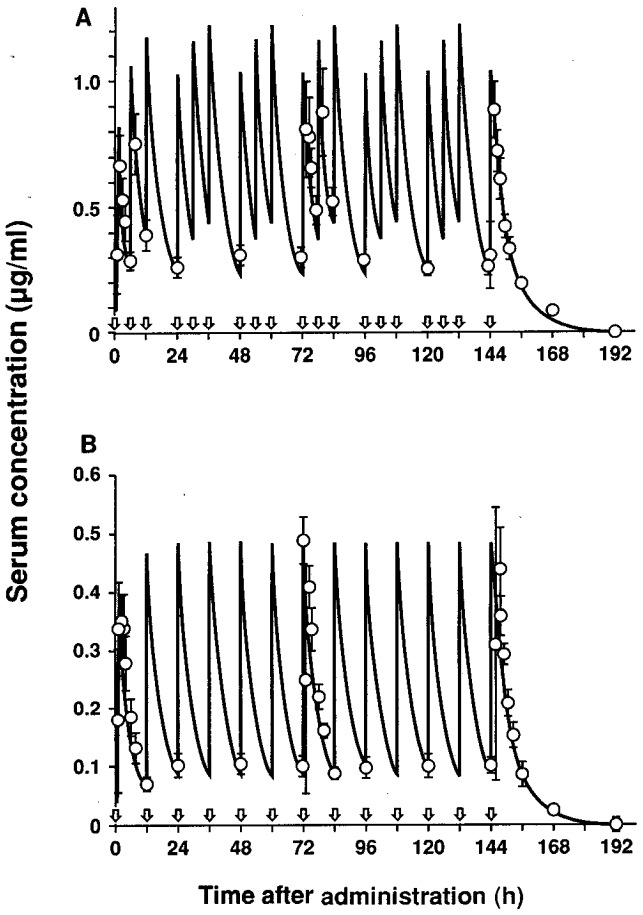

FIG. 5. Observed (○) and predicted (——) serum concentration-time profiles for DU-6859 in six male volunteers after (A) 100 mg t.i.d. for 18 doses on days 1 to 6 plus once on day 7 and (B) 50 mg b.i.d. for 13 doses on days 1 to 7. Times of administration are indicated by arrows. Error bars indicate SD ($n = 6$).

tiple dosing was almost constant, and no change in rate due to consecutive doses was observed. In groups 5 and 6, cumulative urinary excretion was 69.0% ± 6.7% (up to 72 h after the final dose) and 77.0% ± 7.5% of the total dose (up to 48 h after the last dose), respectively. These values were consistent with those found after single oral doses.

DISCUSSION

The dose regimens tested were well tolerated by all volunteers, and all volunteers completed the single- and multiple-dose studies. It has been reported that crystalluria was observed at higher doses of norfloxacin (4) and ciprofloxacin (6). No drug crystals of DU-6859a were noted in the urine following either single or multiple dosing.

DU-6859a as the tablet formulation was found to be rapidly absorbed from the empty gastrointestinal tract, as indicated by the T_{max}s, which were achieved 1.0 to 1.3 h after oral administration. AUC and C_{max} increased linearly over the dose range from 25 to 200 mg. V_{ss}/F ranged from 1.46 to 1.88 liters/kg and clearly exceeded 1 liter/kg, which reflected good tissue penetration for this drug.

Approximately 70% of the dose given was recovered in urine as unchanged drug up to 48 h after a single oral dose, which indicated that the bioavailability of DU-6859a in human volunteers would be at least greater than 70%. Cl_R ranged from 185 to 240 ml/min, and ca. 50% of total drug was bound to serum proteins. Thus, the renal clearance of free drug was much greater than the mean glomerular filtration rate. These data indicated that active processes would take place in the urinary excretion of DU-6859. The high urinary concentrations of unchanged drug were maintained over a long period and represented an important prerequisite for the treatment of urinary tract infections. Nonrenal clearance accounted for about one-third of the Cl_{tot}. Fecal excretion of the drug up to 48 h after the 200-mg dose was only about 3% of the dose. The quantitative contribution of biliary excretion and/or metabolic reactions to this process has not yet been fully investigated.

The effect of food intake on the pharmacokinetics of DU-6859a was investigated by comparing the pharmacokinetic parameters obtained for the single 100-mg dose in fasted and nonfasted states. Food intake slightly increased the MRT, which, however, was not considered clinically significant. All pharmacokinetic parameters other than MRT showed no statistically significant difference between the fasted and nonfasted states ($P > 0.05$, $n = 6$).

During the multiple dosing of 100 mg t.i.d. and 50 mg b.i.d. over 6 days plus once on the 7th day, the measured accumulation of the drug in serum was close to the theoretically predicted value. The cumulative urinary excretion rate in the multiple-dose study was similar to that in the single-dose study. These data indicated that time-dependent effects such as autoinduction or autoinhibition of drug-metabolizing enzymes could be excluded. The peak concentrations after the 100-mg t.i.d. regimen were four to eight times the MIC for 90% of clinically isolated strains such as *Streptococcus pneumoniae*, *Staphylococcus aureus*, and *Klebsiella pneumoniae* (3). After the 50-mg b.i.d. regimen, peak levels were two- to fivefold higher than the MIC for 90% of these strains.

In conclusion, DU-6859a was well tolerated after a single oral dose of up to 200 mg and after 100-mg t.i.d. and 50-mg b.i.d. regimens. DU-6859a was rapidly absorbed from tablet formulation and primarily eliminated by the kidney, with elimination half-lives of 4.4 to 5.0 h. It is expected that DU-6859a will prove to be an effective therapeutic agent in the treatment of various infectious diseases.

ACKNOWLEDGMENTS

We thank Yumi Ohshima and Chiho Ono for expert technical assistance in performing HPLC analyses of the study samples and Miho Nakamura, Sanae Takada, and Michiyuki Kato for performing microscopic urinalysis. The valuable discussions with and support of Keiji Kawamoto, Yasushi Suzuki, and Shohgo Atarashi are gratefully acknowledged.

REFERENCES

1. **Aoki, H., Y. Ohshima, M. Tanaka, O. Okazaki, and H. Hakusui.** 1994. High-performance liquid chromatographic determination of the new quinolone antibacterial agent DU-6859a in human serum and urine using solid-phase extraction with photolysis-fluorescence detection. J. Chromatogr. **660:**365–374.

1a. **Aoki, H., M. Tachibana, M. Tanaka, O. Okazaki, and H. Hakusui.** 1993. DU-6859a, a novel quinolone: identification of the main metabolite in rats, abstr. 1001, p. 302. Program Abstr. 33rd Intersci. Conf. Antimicrob. Agents Chemother. American Society for Microbiology, Washington, D.C.

2. **Okazaki, O., H. Aoki, T. Kurata, and H. Hakusui.** 1991. Metabolic disposition of DU-6859 in rat, abstr. 1508, p. 350. Program Abstr. 31st Intersci. Conf. Antimicrob. Agents Chemother. American Society for Microbiology, Washington, D.C.

3. **Sato, K., K. Hoshino, M. Tanaka, I. Hayakawa, and Y. Osada.** 1992. Antimicrobial activity of DU-6859, a new potent fluoroquinolone, against clinical isolates. Antimicrob. Agents Chemother. **36:**1491–1498.

4. **Swanson, B. N., V. K. Boppana, P. H. Vlasses, H. H. Rotmensch, and R. K.**

Ferguson. 1983. Norfloxacin disposition after sequentially increasing oral doses. Antimicrob. Agents Chemother. **23:**284–288.
5. **Tanaka, M., H. Aoki, T. Kurata, O. Okazaki, and H. Hakusui.** 1993. DU-6859a, a novel quinolone: pharmacokinetics in rats, dogs and monkeys, abstr. 1000, p. 302. Program Abstr. 33rd Intersci. Conf. Antimicrob. Agents Chemother. American Society for Microbiology, Washington, D.C.
6. **Thorsteinsson, S. B., T. Bergan, S. Oddsdottir, R. Rohwedder, and R. Hohm.** 1986. Crystalluria and ciprofloxacin, influence of urinary pH and hydration. Chemotherapy **32:**408–417.
7. **Yamaoka, K., Y. Tanigawara, T. Nakagawa, and T. Uno.** 1981. A pharmacokinetic analysis program (MULTI) for microcomputer. J. Pharmacobio-Dyn. **4:**879–885.