# Pharmacokinetics and Safety of Azthreonam in Healthy Japanese Volunteers

Mitsuyoshi NAKASHIMA*1　Toshihiko UEMATSU*1
Yoshiharu TAKIGUCHI*1 and Yasuyuki MAEDA*2

(Received on December 24, 1984)

*1 Department of Pharmacology, Hamamatsu University School of Medicine
*2 R & D Department, Squibb Japan Inc.

Azthreonam (SQ26, 776), a new, completely synthetic, monocyclic $\beta$-lactam antimicrobial agent, was administered to 29 healthy, Japanese male volunteers by intravenous bolus infusion, intravenous drip infusion, and intramuscular injection as single doses, and intravenous bolus infusion as multiple dose. The pharmacokinetics of single intravenous doses of 500 mg, 1000 mg and 2000 mg were best described by a linear open two-compartment model. The mean peak serum levels were proportional to the doses but the mean serum half-life was essentially independent of the dose administered ($t_{1/2}$=1.5−1.8 h).

The urinary excretion within 24 h ranged from 60-70% of the dose administered as intact azthreonam. Usually, intravenous doses were primarily excreted in the 8 h urine. The pharmacokinetics of an intramuscular dose of 1000 mg fitted a linear open one-compartment model with a mean peak serum level of 66 $\mu$g/ml at 1 h after dosing, and the urinary excretion rate was 80% of the dose administered within 24 h. In the multiple-dose regimen with 1000 mg of azthreonam administered intravenously to six healthy male subjects every 12 h for 5 days, the mean peak serum levels and urinary excretions did not suggest any evidence of drug accumulation. The results of physical examinations and laboratory tests indicated azthreonam was well tolerated with only

*1 浜松医科大学薬理学教室
*2 日本スクイブ株式会社研究開発部
〒150　東京都渋谷区渋谷 2-12-19
長井インターナショナルビル

moderate reversible elevation of CPK values in all subjects at 24 h after intramuscular administration.

**Key words** : azthreonam, SQ26, 776, monobactam, healthy Japanese volunteers, Phase I

Azthreonam is a monocyclic $\beta$-lactam antibiotic with the following formula (Fig. 1) which is produced by chemical synthesis[1]. It has been shown to be active against the majority of aerobic gram-negative bacteria[2].

Pharmacokinetic studies conducted in healthy male volunteers in the U. S. A. showed that azthreonam is well tolerated and displays linear kinetics over an intravenous dose range of 125 to 4000 mg in single dose and 500 and 1000 mg in multiple dose, and an intramuscular dose range of 250 to 1000 mg in single dose and 500 and 1000 mg in multiple dose, with a serum elimination half-life of between 1.3 and 2 h[3)4)5)].

The following report summarizes our single and multiple dose kinetic and safety studies conducted in healthy Japanese subjects.

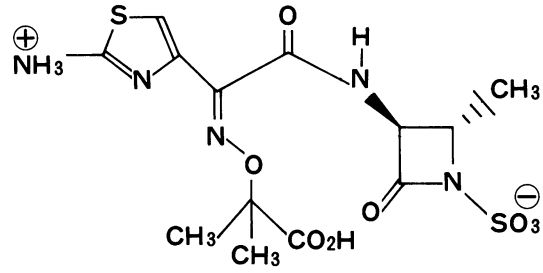


**Fig. 1** Structure of azthreonam (SQ 26, 776).

## Materials and Methods

1 ) Subjects

Twenty-nine healthy male subjects entried into this study after receiving their informed, written consent. All subjects were normal, as judged by physical examinations, chest x-ray, ECG, urinalysis, and serum chemistry profiles (cholesterol, alkaline phosphatase, GOT and GPT). No subjects were accepted with a history of chronic disease or allergy to any drug or other substances. Ages ranged from 22 to 47 years (mean 30.7 years), and weights ranged from 47.0 to 81.0 kg (mean 62.4 kg).

2 ) Administration

Prior to initiation of dosing, subjects were tested for allergy by injecting 400 $\mu$g of azthreonam into the skin of the forearm. For intravenous bolus dosing, 500 mg of azthreonam was administered in 20 ml of sterile water over 3 min, to 3 subjects as single doses, while 1000 mg and 2000 mg of azthreonam was administered in 20 ml of sterile water over 3 min, to 5 subjects in each treatment group as single doses. In the multiple intravenous regimen, 1000 mg of azthreonam was administered nine times at 12-h intervals in 20 ml of sterile water over 2 min to 6 subjects.

For the single 1-h drip infusion, 1000 mg of azthreonam was administered in 250 ml of normal saline to 5 subjects. As the single intramuscular dose, 1000 mg of azthreonam was injected into the right gluteus muscle in 3.5 ml of sterile water to 5 subjects. Approximately 250 ml of water was ingested at the time of drug administration and at 2 and 4 h after dosing to promote urine formation. The subjects were fasted overnight before dosing in the single dose regimens, and fasted before the first, fifth and ninth dosing in the multiple dose regimen.

3 ) Safety evaluation

A series of laboratory tests was performed on each subject before and several times after drug administration as follows : blood pressure, pulse rate, body temperature, respiration rate, and subjective symptoms. Complete blood counts

(hemoglobin, hematocrit, red cell counts, leukocyte counts, platelet counts), serum chemistry profiles (sodium, potassium, chloride, calcium, glucose, urea nitrogen, creatinine, uric acid, cholesterol, total protein, albumin, total bilirubin, alkaline phosphatase, GOT, GPT, LDH, CPK [only for intramuscular dosing]), urinalysis (specific gravity, pH, acetone, sugar, blood, protein, urobilinogen), and ECG were determined before and at 24 and 192 h after injection of the drug in the single-dose regimens, and before and at 48, 120 and 312 h after the first injection of the drug in the multiple-dose regimen.

4 ) Sampling of biological fluids for drug assay

Samples of blood for assay of azthreonam were drawn as follows:

(a) Immediately before drug adminstration and at 1/12, 1/4, 1, 2, 6, 12 and 24 h after completion of the 3-min single-dose intravenous bolus injection,

(b) Immediately before drip infusion and at-1/2, 0, 1/4, 1, 2, 6, 12 and 24 h after completion of single-dose 1-h drip infusion,

(c) Immediately before drug administration and at 1/3, 2/3, 1, 1. 5, 2, 6, 12 and 24 h after completion of single-dose intramuscular injection, and

(d) Immediately before drug administration and at 1/4, 3, 6 and 12 h after completion of the first and ninth doses, at 1/4 and 12 h after completion of the fifth dose, and 24 h after completion of the ninth dose in the multiple-dose intravenous regimen. Serum was prepared for subsequent analysis. Urine samples were obtained before injection of the drug, and cumulative urine collections were made in the intervals of 0 to 2, 2 to 4, 4 to 6, 6 to 8, 8 to 12 and 12 to 24 h in single-dose regimens. Urine samples were also obtained before the first injection and quantitatively complete collections were performed in the intervals of 0 to 4, 4 to 8, and 8 to 12 h after the first and ninth doses, 0 to 12 h after the second, fifth, and sixth doses, and 12 to 24 h after the ninth dose in the multiple-dose regimen.

5 ) Assays and pharmacokinetic methods

Azthreonam concentrations in serum and urine samples were determined by both microbiological methods and high pressure liquid chromatography (HPLC)[5].

A standard two-compartment open model was used to calculate azthreonam pharmacokinetic parameters after intravenous injection. Nonlinear regression analysis was performed, using the computer programs, "Statistical Analysis with Least Squares fitting (SALS)". The following pharmacokinetic parameters were determined by standard methods:

- $V_1$ : volume of distribution of the central compartment
- $V_2$ : volume of distribution of the peripheral compartment
- Vd : apparent volume of distribution
- $k_{12}$ : intercompartmental transfer rate constant
- $k_{21}$ : intercompartmental transfer rate constant
- $k_{10}$ : elimination rate constant
- $t_{1/2}\alpha$ : elimination half-life of $\alpha$-phase
- $t_{1/2}\beta$ : elimination half-life of $\beta$-phase
- AUC: area under the curve based on the trapezoidal rule

A one-compartment model was used to calculate the pharmacokinetic parameters after intramuscular injection in a manner similar to that just described for the intravenous study.

## Results

1 ) Intravenous bolus pharmacokinetics

The bioassay and the HPLC assay for azthreonam in serum and urine samples gave equivalent

results. The results of the bioassay will be described in detail below.

Fig. 2 shows the mean serum concentraions of azthreonam after single 500 to 2000 mg intravenous bolus doses. The mean peak levels at 5 min after 500, 1000 and 2000 mg doses were 70.7, 130.6 and 256.0 $\mu$g/ml, respectively, and proportional to dose.

The urinary excretion of azthreonam in each dose is shown in Tab. 1. Unchanged azthreonam equivalent to 72.2%, 57.3% and 73.7% of the 500, 1000 and 2000 mg doses respectively was excreted within 24 h, primarily within 8 h. The serum pharmacokinetics of azthreonam administered intravenously were described by an open, linear two-compartment model ; the kinetic parameters are shown in Tab. 2.

2 ) Intravenous drip infusion pharmacokinetics

Fig. 3 shows the mean serum concentration of azthreonam after intravenous drip infusion of a single 1000 mg dose. Mean peak level at the end of infusion was 93.4 $\mu$g/ml. Urinary excretion is shown in Tab. 1. A total of 59.3% of the dose was excreted unchanged within 24 h, mostly within 8 h.

3 ) Intramuscular pharmacokinetics

Fig. 4 shows the mean serum concentration of azthreonam after a single 1000 mg intramuscular dose. The mean peak level, attained at about 40 min, was 66.3 $\mu$g/ml. The serum pharmacokinetics of azthreonam administered intramuscularly were described by an open linear one-compartment model (Tab. 3). The urinary excretion is shown in Tab. 1. A total of 81.3% of the dose was excreted unchanged within 24 h, mostly

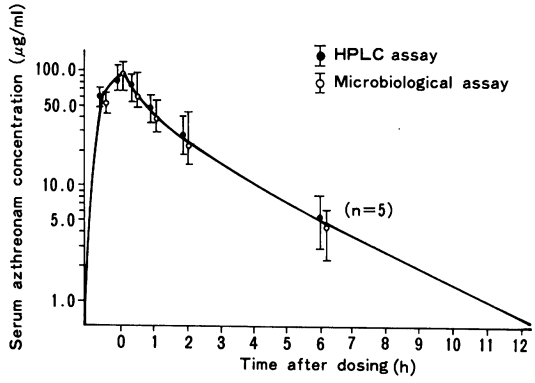


Fig. 3 Mean serum concentration of azthreonam after 1-h intravenous drip infusion of 1000 mg.

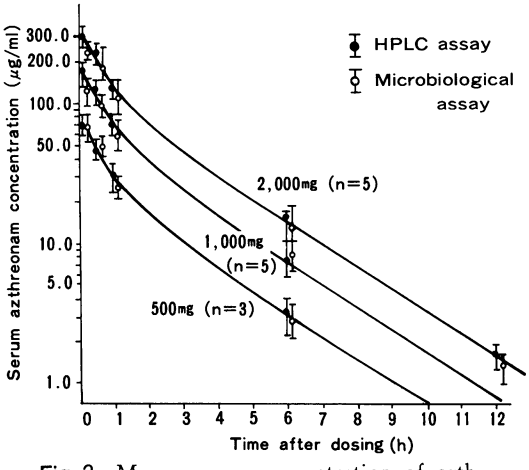


Fig. 2 Mean serum concentration of azthreonam after 3-min intravenous bolus infusion.

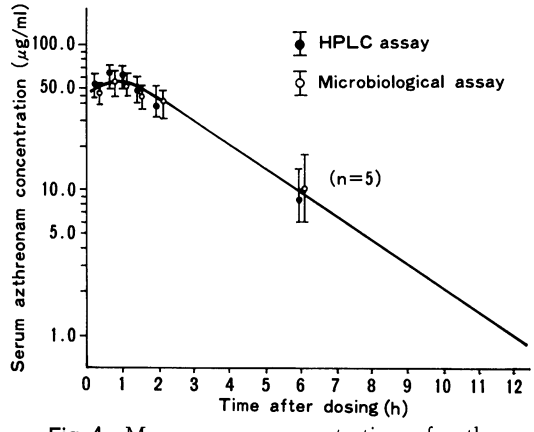


Fig. 4 Mean serum concentration of azthreonam after intramuscular injection of 1000 mg.

Tab. 1 Mean Urinary Excretion of Azthreonam after Single Intravenous and Intramuscular Administration

| dose (mg) | interval(h) after dosing | urinary concn. (μg/ml) | azthreonam excreted(mg) | mean urinary recovery(%) |
|---|---|---|---|---|
| 500 (i.v.) | 0 - 2 | 2,150 ( 2950 ~ 1600 ) | 222.8 (254.4 ~ 191.8) | |
| | 2 - 4 | 335 ( 610 ~ 190 ) | 77.6 ( 89.6 ~ 61.0) | |
| | 4 - 6 | 155 ( 225 ~ 120 ) | 34.3 ( 40.3 ~ 30.8) | |
| | 6 - 8 | 113 ( 145 ~ 58 ) | 16.0 ( 18.9 ~ 13.1) | 0 - 8h 70.1 |
| | 8 -12 | 26.5 ( 36 ~ 21 ) | 8.4 ( 10.4 ~ 7.0) | |
| | 12 -24 | 2.3 ( 3.0~ 1.8) | 1.7 ( 2.3 ~ 1.4) | 0 -24h 72.2 |
| 1000 (i.v.) | 0 - 2 | 1,350 ( 2630 ~ 330 ) | 315.7 (440.0 ~ 218.3) | |
| | 2 - 4 | 973 ( 2950 ~ 260 ) | 133.8 (236.0 ~ 80.6) | |
| | 4 - 6 | 319 ( 940 ~ 79.5) | 59.5 (110.0 ~ 37.6) | |
| | 6 - 8 | 116 ( 259 ~ 39.5) | 29.1 ( 37.6 ~ 21.4) | 0 - 8h 53.8 |
| | 8 -12 | 36.3 ( 60.0~ 21.0) | 26.9 ( 32.5 ~ 20.3) | |
| | 12 -24 | 13.6 ( 20.3~ 5.3) | 8.5 ( 21.3 ~ 2.8) | 0 -24h 57.3 |
| 2000 (i.v.) | 0 - 2 | 5,470 (10700 ~ 2350 ) | 730.4 (930.9 ~ 493.5) | |
| | 2 - 4 | 2,314 ( 3800 ~ 820 ) | 373.1 (562.5 ~ 274.7) | |
| | 4 - 6 | 2,532 ( 4100 ~ 1380 ) | 215.1 (368.0 ~ 122.8) | |
| | 6 - 8 | 1,030 ( 1250 ~ 530 ) | 96.3 (177.5 ~ 63.8) | 0 - 8h 70.7 |
| | 8 -12 | 239 ( 430 ~ 96 ) | 44.1 ( 51.5 ~ 34.7) | |
| | 12 -24 | 38.4 ( 48.0~ 21.5) | 15.6 ( 17.4 ~ 13.6) | 0 -24h 73.7 |
| 1000 (d.i.) | 0 - 2 | 1,540 ( 3150 ~ 250 ) | 308.2 (582.8 ~ 52.5) | |
| | 2 - 4 | 1,463 ( 2500 ~ 375 ) | 146.7 (247.2 ~ 70.4) | |
| | 4 - 6 | 436 ( 1180 ~ 92.5) | 83.2 (123.4 ~ 39.0) | |
| | 6 - 8 | 208 ( 530 ~ 105 ) | 26.9 ( 39.2 ~ 14.7) | 0 - 8h 56.5 |
| | 8 -12 | 76.2 ( 220 ~ 7.1) | 21.6 ( 40.1 ~ 5.8) | |
| | 12 -24 | 12.4 ( 21.5~ 5.3) | 6.1 ( 8.4 ~ 1.9) | 0 -24h 59.3 |
| 1000 (i.m.) | 0 - 2 | 2,228 ( 4600 ~ 660 ) | 280.8 (340.4 ~ 195.4) | |
| | 2 - 4 | 3,071 ( 8025 ~ 1210 ) | 266.5 (385.2 ~ 167.5) | |
| | 4 - 6 | 1,387 ( 1970 ~ 660 ) | 160.4 (324.9 ~ 78.8) | |
| | 6 - 8 | 480 ( 850 ~ 250 ) | 63.1 (116.0 ~ 26.5) | 0 - 8h 77.1 |
| | 8 -12 | 186 ( 420 ~ 40 ) | 35.9 ( 58.4 ~ 16.9) | |
| | 12 -24 | 17.9 ( 36.2~ 8.7) | 6.4 ( 7.9 ~ 2.5) | 0 -24h 81.3 |

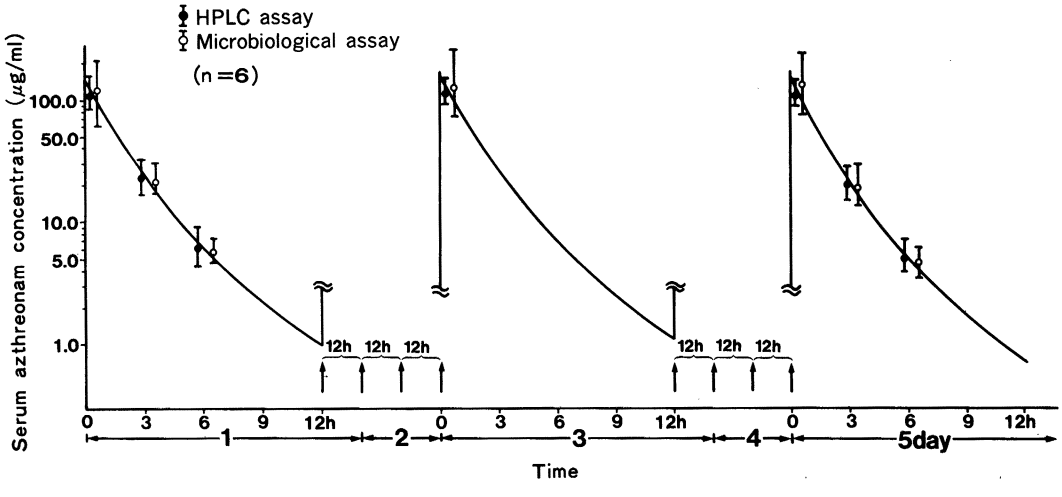


Fig. 5 Mean serum concentration of azthreonam during 2-min intravenous bolus infusion of 1000 mg q 12 h for 5 days.

within 8 h.

4 ) Multiple-dose pharmacokinetics

Fig. 5 shows the mean serum concentration of azthreonam after administration of multiple 1000 mg intravenous doses. The mean peak levels at 15 min after the first, fifth and ninth administrations were 117. 2, 141. 8 and 142. 3 $\mu$g/ml, respectively. The values for serum trough levels over 5 days indicated a lack of detectable accumulation of azthreonam during the q 12 h dosage regimens. The urinary excretion rates within 12 h after the first, second, fifth, sixth and ninth administrations were 80. 7, 70. 0, 79. 1, 80. 0 and 83. 0%, respectively. The urinary excretion pattern after the first administration was very similar to that after the ninth administration.

5 ) Safety

Human tolerance of single- or multiple-intravenous and intramuscular doses of azthreonam was acceptable.

(a) Physiological function tests : One subject developed mild fever 4 h after completion of drip infusion, but the body temperature returned to normal by the next morning without treatment. No fever was observed in the second follow-up trial in the same subject administered 500 mg in a single infusion one month later. No significant change was observed in any other subjects.

(b) Subjective symptoms : One subject developed slight stomatitis at the site of the gingiva of the stomach tooth 24 h after the final dose in the multiple-dose regimen. The symptom disappeared by the next day without treatment. Three subjects in the multiple-dose group experienced transient, mild taste alterations at every dosing from the third to the last infusion. Otherwise, they complained of no other unpleasant feeling due to azthreonam. In the intramuscular group, only one subject complained of mild pain at the time of intramuscular injection. Neither rubor nor edema were observed at injection sites.

change after drug administration and no eosino-

(c) Laboratory tests : No significant change was observed in the laboratory tests except for mild, reversible elevation of CPK values which was observed in all subjects receiving intramuscular injection. The elevation of CPK values was not associated with any symptom of physical findings and returned to normal by the follow-up examination one week after the dosing.

## Discussion

Azthreonam is highly active against aerobic gram-negative organisms, but lacks significant activity against gram-positive organisms[7].. The minimal inhibitory concentration (MIC) values against aerobic gram-negative organisms were equal, if not superior, to those of aminoglycosides and the third-generation cephalosporins[8][9]. And it was also reported that this drug was efficacious in infections due to aerobic gram-negative organisms[10]. If azthreonam proves to have a more favorable safety profile than the aminoglycosides, it is expected to be a more useful chemotherapeutic agent in human.

The purpose of this study was to examine whether the pharmacokinetic and safety profiles of azthreonam in Japanese are similar to those reported for Americans.

The mean serum concentrations of azthreonam after single intravenous doses in our study were very similar to those reported by Swabb et al.[3]. The pharmacokinetic parameters in the single intravenous regimens, therefore, are essentially same as those reproted for Americans. The cumulative urinary excretion in our study ranged from 57. 3% to 73. 7% of the doses administered and that reported for Americans was 68% of single intravenous doses.

For the single intramuscular dose of 1000 mg,

Tab. 2 Pharmacokinetics of Azthreonam after 3-min Intravenous Bolus Infusion

| Parameters \ Dose | | 500mg(n=3) | 1,000mg(n=5) | 2,000mg(n=5) |
|---|---|---|---|---|
| A | ($\mu$g/ml) | 50.2 | 82.7 | 153 |
| B | ($\mu$g/ml) | 31.4 | 76.5 | 147 |
| $\alpha$ | ($h^{-1}$) | 2.61 | 4.58 | 3.46 |
| $\beta$ | ($h^{-1}$) | 0.394 | 0.375 | 0.424 |
| $t_{1/2\alpha}$ | (h) | 0.266 | 0.151 | 0.200 |
| $t_{1/2\beta}$ | (h) | 1.76 | 1.85 | 1.63 |
| $K_{12}$ | ($h^{-1}$) | 0.930 | 1.84 | 1.21 |
| $K_{21}$ | ($h^{-1}$) | 1.25 | 2.39 | 1.91 |
| $K_{10}$ | ($h^{-1}$) | 0.825 | 0.716 | 0.768 |
| AUC | ($\mu$g·h/ml) | 99.0 | 222 | 389 |
| $V_1$ | ($\ell$) | 6.12 | 6.28 | 6.69 |
| $V_2$ | ($\ell$) | 4.57 | 4.83 | 4.21 |
| $Vd_{(extrap)}$ | ($\ell$) | 15.9 | 13.1 | 13.6 |

Tab. 3 Pharmacokinetics of Azthreonam after Intramuscular Injection of 1000 mg

| Kel($h^{-1}$) | $t_{1/2}$(h) | AUC($\mu$g·h/ml) | $C_{max}$($\mu$g/ml) | $t_{max}$(h) |
|---|---|---|---|---|
| 0.344 | 2.01 | 229.71 | 66.3 | 0.333~1 |

the mean serum concentrations in our study were relatively higher than those of Americans[3]. This fact led to the discrepancy in the mean AUC values between Japanese subjects (230 $\mu$g·hr/ml) and Americans (180 $\mu$g·hr/ml). The values for distribution and elimination parameters, k el and $t_{1/2}$ el, after intramuscular injection to Japanese were similar to those reported for Americans. The mean cumulative urinary excretion of azthreonam in our study was 81.3% of the dose, while that found in American subjects was 63% of the dose.

Both our intravenous multiple-dose regimen (q 12 h for 5 days) and that for Americans (q 8 h for 7 days)[4] indicated no evidence of drug accumulation in serum concentrations or in cumulative urinary excretions.

In the drip infusion study, one subject developed transient mild fever 4 h after the end of drip infusion. This subject had slight sore throat and headache in the morning, but did not complain thereof before dosing. WBC counts did not change after drug administration and no eosinophilia was observed. We considered this fever to be due to a common cold rather than to azthreonam. To confirm this consideration, we challenged the follow-up test by half of the dose of the previous drip infusion study after receiving the subject consent. No fever was observed in that trial.

Transient, mild taste alteration was complained by three of the 29 subjects. Swabb et al. reported the same symptom in 4 of 18 subjects[4]. In all subjects received intramuscular injection the CPK values elevated reversibly, but only one subject complained of mild pain at the injection site. There was neither phlebitis nor inflammation. The safety profile of azthreonam in intramuscular use remains to be investigated.

## References


1) Sykes, R. B., Cimarusti, C. M., Bonner, D. P. et al. : Monocyclic $\beta$-lactam antibiotics produced by bacteria. Nature (London), 291 : 489-491 (1981).
2) Sykes, R. B., Bonner, D. P., Bush, K. et al. : Azthreonam (SQ26,776), a synthetic monobactam specifically active against aerobic gram-negative bacteria. Antimicrob. Agents Chemother., 21 : 85-92 (1982).
3) Swabb, E. A., Sugerman, A. A., Platt, T. B. et al. : Single-dose pharmacokinetics of the monobactam azthreonam (SQ26,776) in healthy subject. ibid., 21 : 944-949 (1982).
4) Swabb, E. A., Sugerman, A. A. and McKinstry, D. N. : Multiple-dose pharmacokinetics of the monobactam azthreonam (SQ26, 776) in healthy subjects. ibid., 23 : 125-132 (1983).
5) Scully, B. E., Swabb, E. A. and Neu, H. C. : Pharmacology of azthreonam after intravenous infusion. ibid., 24 : 18-22 (1983).
6) Pilkiewicz, F. G., Remstung, B. J., Fisher, S. M. et al. : High-pressure liquid chromatographic analysis of azthreonam in sera and urine. ibid., 23 : 852-856 (1983).
7) Sykes, R. B., Bonner, D. P., Bush, K. et al. :

Monobactams-monocyclic β-lactam antibiotics produced by bacteria. J. Antimicrobial Chemother., 8 (Suppl. E) : 1-16 (1981).
8) Wise, R., Andrews, J. M. and Hancox, J. : SQ26,776, a novel β-lactam : an in-vitro comparison with other antimicrobial agents. ibid., 8 (Suppl. E) : 39-47 (1981).
(Suppl. E) : 39-47 (1981).
9) Neu, H. C. and Labthavikul, P. : Antibacterial activity of a monocyclic β-lactam SQ26,776. ibid., 8 (Suppl. E) : 111-122 (1981).
10) Bonner, D. P., Whitney, C. O., Baughn, B. H. : In-vivo properties of SQ26,776. ibid., 8 (Suppl. E) : 123-130 (1981).