Pharmacokinetics of a New Parenteral Oligosaccharide Antibiotic, SCH27899 (Ziracin), in Healthy Subjects

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The pharmacokinetic properties of an everninomicin antibiotic (SCH27899; Ziracin) were studied with healthy Japanese male volunteers by single (1, 3, 6, and 9 mg/kg of body weight) and multiple 60-min intravenous infusions (3, 6, and 9 mg/kg once daily for 10 consecutive days following a 2-day interval after the initial dose). At single doses the peak serum concentrations and the area under the serum concentration-time curve linearly increased with the dose. While total body clearance (CL; 31.2 to 45.6 ml/kg/h) and percent cumulative urinary recovery as unchanged drug (4.9 to 7.1%) were rather constant irrespective of doses, the terminal half-life of $\gamma$ phase ($t_{1/2\gamma}$; 14.2 to 19.6 h) were slightly prolonged at the higher two doses compared with the lower two doses. With repeated doses of SCH27899, a statistically significant decrease and increase were found in CL and $t_{1/2\gamma}$, of about 36% and 21%, respectively, although these changes probably be clinically irrelevant. The most commonly reported adverse events were local reactions such as erythema, pain, and palpable venous cord of mild to moderate degree around the injection site, which could be managed by changing the injection sites.

**MATERIALS AND METHODS**

**Volunteers.** Before the implementation of this study, the research protocol and the consent form were reviewed and approved by the Ethics Committee of Shiotoro Clinic, 5332-1 Shiotoro-cho, Hamamatsu, Japan. Volunteers were selected on the basis of physical examination, medical history, and screening laboratory tests. Sixty-three healthy male subjects aged 24.1 ± 4.0 (mean ± standard deviation [SD]; range, 20 to 37) years and weighing 64.0 ± 8.0 (51.4 to 79.4) kg participated in the study after giving informed consent. Each subject received only one treatment regimen with a SCH27899 or placebo infusion.

**Study protocol.** The primary objective of this study was to evaluate the safety and tolerability of SCH27899. With six volunteers receiving treatment in each treatment group, there was an 80% chance of at least one occurrence of any untoward event with an incidence rate of 25%.

The pharmacokinetics and safety were first examined by single intravenous administrations of SCH27899 (1, 3, 6, and 9 mg/kg of body weight) in a dose-escalating manner. At each dose, six and three subjects received SCH27899 dissolved in dextrose and placebo (dextrose only), respectively, by a single-blind method. Venous blood samples (5 ml) were collected before (0 h) and 0.5, 1, 1.5, 2, 4, 6, 8, 12, 24, 48, 72, and 144 h after the start of 60-min intravenous infusion. Urine was collected as voided just before administration, which served as a blank for measurement, and at intervals of 0 to 4, 4 to 8, 8 to 12, 12 to 24, and 24 to 48 h after the beginning of drug administration. The repeated-dose study (3, 6, and 9 mg/kg) was also conducted in a dose-escalating manner. At each dose, six and three subjects received a single 60-min infusion of SCH27899 dissolved in dextrose and of placebo (dextrose only), respectively, by a single-blind method on day 1. After a 2-day interval, the same treatment was administered once daily for 10 consecutive days from day 4 to day 13. Venous blood samples were also obtained in the same manner as in the single-dose study on days 1 and 13. Additionally, blood was withdrawn from each subject just before each dose, at the end of infusion on days 5 and 8, and 7 days after the last administration. Urine was collected in the same manner as in the single-dose study on days 1 and 13 and additionally as 24-h block samples on the other dosing days and for 3 days after the last administration.

All subjective and objective symptoms either observed by the investigators or reported by the subject spontaneously or in response to a direct question were recorded. If any adverse event occurred after dosing, the subject was followed with appropriate treatment and close medical supervision. Casually and severity ratings were determined. In the single-dose study, blood biochemistry and hematology tests, urinalysis, and an electrocardiogram were performed at screening, before administration, and 24 h, 7 days, and 14 days after administration. Vital signs including body temperature and blood pressure were monitored just before and periodically up to 48 h after administration. In the multiple-dose

Grum-positive cocci have reemerged as important nosocomial pathogens around the world, especially in the last decade (8). To combat increasing penicillin resistance among pneumococci and virulent streptococci, active agents against such pathogens are increasingly needed, particularly under the threat of the emergence of those resistant to vancomycin and structurally related teicoplanin (3, 11, 13). SCH27899, O-(1R)-4-O-(2,4-dihydroxy-6-methylbenzoyl)-2,3-O-methylened-xylopyranosylidene-(1→3)-4-O-methyl-3-nitro-$\alpha$-L-arabinopyranosyl-(1→3)-3-O,2,6-dideoxy-4-O-[3,5-dichloro-4-hydroxy-2-methoxy-6-methylbenzoyl]-$\beta$-D-arabinopyranosyl-(1→4)-O,2,6-dideoxy-D-arabinopyranosylidene-(1→3)-4-O,2,6-dideoxy-3-C-methyl-$\beta$-D-mannopyranosyl-(1→3)-3,4-O,6-deoxy-3-C-methyl-$\beta$-D-mannopyranosyl-(1→3)-3,4-O,6-deoxy-4-O-methyl-$\beta$-galactopyranosyl(1→4)-2,6-di-O-methyl-$\beta$-mannopyranoside (Ziracin), is an oligosaccharide, everninomicin antibiotic with activity primarily against gram-positive pathogens including glycopeptide-resistant enterococci, oxacillin-resistant staphylococci, and penicillin-resistant streptococci and pneumococci (6).

In the present study the pharmacokinetics and tolerability of SCH27899 were studied with healthy Japanese male volunteers to obtain information to guide the rational use of this agent for the patients. This paper reports that it was possible at proposed clinical doses to attain serum concentrations high enough to exceed the MICs and MIC90s for various clinical isolates (7, 10, 12), including multiresistant staphylococci and enterococci, and that safety will be maintained for patients at those doses.

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study, blood biochemistry and hematology tests, urinalysis, and an electrocardiogram were performed at screening, before dosing, and 24 h after dosing on days 1, 2, 4, 7, and 10, and 7 days after the end of dosing. Vital signs were monitored just before the first administration and periodically up to 48 h after the last administration.

Assays. The 4-ml blood samples collected in the course of the study were allowed to stand at room temperature for 30 min and then centrifuged at 1,500 × g for 15 min at 4°C. The separated sera were transferred into plastic tubes. The volume of time-block urine samples was measured, and 0.5 ml of 1 M sodium dihydrogenophosphate (pH 6.9) and 5 ml of acetonitrile were added to urine aliquots (5 ml). These serum and buffered urine samples were stocked at 0 to −20°C until analyzed. Serum and urinary concentrations of SCH27899 were determined by validated high-performance liquid chromatography (HPLC) using thiabendazole as an internal standard (IS). An aliquot (250 μl) of serum, calibration standard, or quality control (QC) sample was well mixed with 50 μl of IS solution in methanol (5 μg/ml) in a centrifugation tube to which was added 500 μl of acetonitrile. The mixture was centrifuged at approximately 2,700 × g for 10 min at 20°C. The supernatant was transferred to another tube and the solvent was evaporated under nitrogen gas flow at about 40°C. The residue was reconstituted with 200 ml of 50 mM sodium dihydrogenophosphate (pH 6.9) and acetonitrile (1:1, vol/vol). The resolubilization was followed by centrifugation at approximately 4,000 × g for 5 min at room temperature. An aliquot (75 μl) of the supernatant was injected onto an HPLC. For urine, the calibration curve was prepared using blank human urine, 1 M sodium dihydrogenophosphate, and acetonitrile (100:1:100, vol/vol/vol) supplemented with 200 to 50,000 ng of SCH27899/ml. The QC samples were prepared at concentrations of 200, 20,000, and 40,000 ng of SCH27899/ml. To an aliquot (1 ml) of buffered urine, calibration standard, or QC sample, 50 μl of IS in methanol (25.0 μg/ml) was added. After agitation, the mixture was centrifuged at approximately 4,000 × g for 5 min at room temperature. An aliquot (75 μl) of the supernatant was injected onto the HPLC. The HPLC system (Hitachi, Tokyo, Japan) consisting of a pump (L-7100), an autosampler (L-7200), and a UV detector (L-7400; wavelength, 300 nm) was operated at ambient temperature. The flow rate was 1.0 ml/min. An octyldecyl silane analytical column (Asahipak OD-P5; 4.6 mm ID by 150 mm) was used. The mobile phase was a mixture of 20 mM sodium dihydrogenophosphate (pH 7.8) and acetonitrile (31.21, vol/vol). The solution was filtered through a membrane filter (pore size: 0.45 μm) and degassed before use. The HPLC system was operated at ambient temperature. The flow rate was 1.0 ml/min.

The calibration curve for serum assay was prepared ranging to 50,000 ng of SCH27899/ml. The HPLC peak height ratio (SCH27899/IS, y) versus SCH27899 concentration (x) data from the calibration curve constructed were evaluated using a linear equation for each calibration curve (y = ax + b) by weighed (1/x) least-squares regression for both serum and urine samples. The lower limit of quantitation was established at 50 and 200 ng/ml for serum and urine samples, respectively. The QC samples were prepared at concentrations of 150, 25,000, and 40,000 ng/ml for serum, the within-day precision (% coefficient of variation) ranged from 1.6 to 5.3% at the three concentrations (0.150, 4.00, and 40.0 μg/ml) on all three validation days (days 1, 2, and 3), and the between-day precision values obtained for the samples of these concentrations were 13.8, 4.7, and 4.2, respectively, on five different days. In urine, the within-day precision ranged from 0.9 to 3.7% at three concentrations (0.600, 6.0, and 40.0 μg/ml) on all three validation days, and the between-day precision values were 2.6, 3.7, and 4.6%, respectively, on three different days. Results for the standards and QC samples met the criteria for acceptable performance of the method during the period of study sample analysis.

Pharmacokinetic and statistical analyses. As compared to a two-compartment open model, serum concentrations of SCH27899 apparently fitted much better to a three-compartment open model, especially around the peak concentration and open model, serum concentrations of SCH27899 apparently fitted much better to a three-compartment open model, especially around the peak concentration and open model, serum concentrations of SCH27899 apparently fitted much better to an acceptable performance of the method during the period of study sample analysis.

In the single-dose study, SCH27899 was well tolerated with no injection-site reactions. With repeated administrations of 3 mg of SCH27899/kg, such injection site reactions as erythema, pain, and palpable venous cord of mild to moderate degree were reported from daily 5 by three of six subjects. However, these local reactions could be avoided or managed by changing the injection sites, so that the final incidence of local reactions was three of six, one of six, and two of six subjects at the doses of 3, 6, and 9 mg/kg, respectively. In a subject in the 6 mg/kg repeated-dose group, the drug was discontinued during the sixth consecutive dose because of the appearance of a skin rash with itching of moderate degree near the end of infusion. Immediately after the discontinuation of drug infusion, the skin rash started to rapidly disappear. This subject was excluded from the final data analysis. Desquamation of the fingertips of mild degree was noted in two of six subjects after the completion of the highest repeated doses.

DISCUSSION

The present study revealed pharmacokinetic properties of SCH27899 in healthy subjects which support the feasibility of once-daily administration for clinical practice.
In the single-dose study, the C_{1h} and AUC increased in proportion to the dose within the dose range examined (1 to 9 mg/kg; Fig. 2). The percent cumulative urinary recovery as unchanged drug was less than 10% irrespective of dose, showing that the main route of excretion of SCH27899 is extrarenal, through biotransformation by the liver. At the lowest dose the terminal (\( \gamma \)) phase of serum concentration could not be fully described due to the methodological imbalance between the actual concentration and the detection limit for measurement. In fact, an attempt for a subject of the lowest dose group to fit his serum concentrations of SCH27899 by a three-compartment open model failed with no convergence because of the lack of information on the terminal phase. As judged from the above finding together with the observation that the CL remained almost unaltered within the dose range of 3 to 9 mg/kg, the possibility that the hepatic capacity to biotransform this agent might be saturated around the present dose range would not be the case, although the \( t_{1/2\gamma} \) was significantly prolonged at the higher two doses compared with the lower two doses.

On the other hand, in association with repeated administrations of SCH27899, a statistically significant decrease and increase were found in the CL and \( t_{1/2\gamma} \) of about 36 and 21%, respectively, resulting in a significant increase in the AUC of about 17%. Although these changes may be clinically irrelevant, precaution should be taken against some possible abnormal accumulation of SCH27899 in the body with repeated administrations, especially in patients with impaired hepatic function.

There were no clinically significant dose-related changes in hepatic or renal function tests or clinical assessments that might hinder further clinical development. However, the injection-site reactions of mild to moderate degree and the adverse events of skin rash and fingertip desquamation, which might be due to some allergic reactions and should be carefully investigated in further study, should urge us to assess the ratio of risk to benefit of the actual use of this agent in patients.

The glycopeptide antibiotic vancomycin and the structurally related antibiotic teicoplanin have been considered to be the last lines of defense against a variety of serious infections caused by gram-positive organisms such as enterococci and staphylococci (9, 11). The recent rapid emergence and spread of vancomycin-resistant enterococci (VRE) is, therefore, of grave concern to both medical practitioners and their patients. Reports of the extensive spread of vancomycin-resistant, i.e., heteroresistant, MRSA (methicillin-resistant \( \text{Staphylococcus aureus} \)) strains in Japanese hospitals after 30 years of vancomycin use as the drug of choice for the treatment of MRSA are

![FIG. 1. Time profile of the serum concentration of SCH27899 after single intravenous 60-min infusions of 1 (●), 3 (■), 6 (▲), and 9 (○) mg/kg.](image-url)
of particular concern in light of observations that the high-level VanA vancomycin resistance gene can be transferred from enterococci to staphylococci in vitro (1). To reduce the spread of VRE and other multiresistant bacteria such as heteroresistant MRSA and drug-resistant Streptococcus pneumoniae, the clinical development of safe and effective innovative agents should be promoted and an aggressive infection and antibiotic control strategy established (3). For example, the development of new glycopeptides, including LY264826 and its semisynthetic derivative LY333328, is of special interest because of their potential usefulness in the treatment of VRE infections (2). However, these agents are not necessarily free from potential cross-resistance with vancomycin and teicoplanin.

SCH27899 is structurally different from vancomycin or teicoplanin.
the C1h exceeds these MIC and MIC 90 values. As seen from 12), it is one of the essential factors for clinical utilization that from 0.015 to 1.0 SCH27899 for most gram-positive strains of clinical isolates, was also noted at steady state. present study of SCH27899, a similar decrease in CL of 36% comycin could display a significant metabolism (4). In the in hepatic biotransformation in light of some reports that van-
sively through the renal route, this is explained by the decrease (14). Although vancomycin is considered to be excreted exclu-
vosin in patients with impaired renal function. A decrease in CL of around 30% has been reported for vancomycin at steady state as compared with initial therapy (14). Although vancomycin is considered to be excreted exclu-
ively through the renal route, this is explained by the decrease in hepatic biotransformation in light of some reports that van-
comycin could display a significant metabolism (4). In the present study of SCH27899, a similar decrease in CL of 36% was also noted at steady state. Since preclinical studies indicate that the MICs of SCH27899 for most gram-positive strains of clinical isolates, including multiresistant staphylococci and enterococci, range from 0.015 to 1.0 µg/ml with MICC90S of 0.12 to 0.5 µg/ml (7, 10, 12), it is one of the essential factors for clinical utilization that the C1h exceeds these MIC and MIC90 values. As seen from Fig. 3, the plasma concentration of SCH27899 above around 1.0 µg/ml could be maintained for 12 to 48 h by the adminis-
tration of 3 to 9 mg/kg, depending on the dose. Vancomycin is reported to have a postantibiotic effect (PAE) of 2 to 3 h against S. aureus (5), and teicoplanin is shown to have an even longer PAE than vancomycin (16). SCH27899 is also consid-
ered to have a PAE of a little longer than vancomycin (unpub-
lished observation). Therefore, on the basis of the present observations on pharmacokinetics and safety, the once-daily regimens of 3 to 9 mg of SCH27899/kg should be tested in further clinical investigation even against multiple-resistant gram-positive bacterial infections. This study was carried out using healthy male Japanese volunteers only, and more data may be needed for treatment of actual patients or females, especially to explore the relationship between serum concentra-
tion and clinical efficacy.

PLATE 1. Pharmacokinetic parameters after repeated intravenous 60-min infusions of SCH27899

<table>
<thead>
<tr>
<th>Dose (mg/kg)</th>
<th>Day</th>
<th>C1h (µg/ml)</th>
<th>t1/2 (h)</th>
<th>AUC (µg · h/ml)</th>
<th>V1/2 (ml/kg)</th>
<th>CL (ml/h)</th>
</tr>
</thead>
<tbody>
<tr>
<td>3</td>
<td>1</td>
<td>31.9 ± 2.4</td>
<td>0.507 ± 0.104</td>
<td>1.37 ± 0.23</td>
<td>14.5 ± 1.4</td>
<td>80.6 ± 11.1</td>
</tr>
<tr>
<td></td>
<td>13</td>
<td>35.1 ± 3.6</td>
<td>0.415 ± 0.069</td>
<td>1.29 ± 0.09</td>
<td>17.9** ± 0.9</td>
<td>112.4*** ± 18.2</td>
</tr>
<tr>
<td>6</td>
<td>1</td>
<td>58.1 ± 3.7</td>
<td>0.456 ± 0.069</td>
<td>1.43 ± 0.11</td>
<td>16.6 ± 0.6</td>
<td>152.7 ± 9.6</td>
</tr>
<tr>
<td></td>
<td>13</td>
<td>62.9 ± 5.2</td>
<td>0.477 ± 0.090</td>
<td>1.48 ± 0.07</td>
<td>20.2** ± 1.3</td>
<td>211.0 ± 34.9</td>
</tr>
<tr>
<td>9</td>
<td>1</td>
<td>91.9 ± 9.1</td>
<td>0.453 ± 0.152</td>
<td>1.53 ± 0.22</td>
<td>16.6 ± 1.2</td>
<td>251.1 ± 31.5</td>
</tr>
<tr>
<td></td>
<td>13</td>
<td>92.6 ± 9.8</td>
<td>0.302 ± 0.116</td>
<td>1.31 ± 0.18</td>
<td>19.9*** ± 1.0</td>
<td>338.7**ed ± 35.3</td>
</tr>
</tbody>
</table>

a Results by three-compartment open model analysis.
b One subject, for whom the administration was discontinued due to an adverse event, was excluded from analysis.
c Comparison between the first (day 1) and last (day 13) doses by Student’s paired t test, * P < 0.05; ** P < 0.01; *** P < 0.001.
d Comparison between AUC0–24 after the first dose (day 1) and AUC0–24 after the last dose (day 13) by Student’s paired t test, * P < 0.05; ** P < 0.01; *** P < 0.001.

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