Pharmacokinetic Interaction of Zonisamide in Rats. Effect of Other Antiepileptics on Zonisamide

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The pharmacokinetics of zonisamide (ZNS) and the effects of phenobarbital (PB), valproic acid (VPA), carbamazepine (CBZ) and phenytoin (PHT) on ZNS kinetics were investigated in rats. The effects of other antiepileptics on the serum protein binding, erythrocyte distribution and metabolism of ZNS were also studied in vitro to elucidate the mechanism of pharmacokinetic interaction of ZNS. ZNS showed a linear disposition kinetics after oral administration of ZNS within the dose examined. Moreover, the pharmacokinetic behaviors of ZNS were not altered after multiple dosing. The decreased $t_{1/2}$ value of ZNS by PB or CBZ pretreatment and the increased $Vd/F$ value of ZNS by VPA pretreatment were observed, although it showed no marked effect of PHT on ZNS kinetics. The enhanced metabolism of ZNS was observed by PB or CBZ pretreatment from an in vitro metabolism study. The serum protein binding and erythrocyte distribution of ZNS showed no significant change in the presence of other antiepileptics in vitro. These results indicate that the decreased $t_{1/2}$ value of ZNS is attributable to the enzyme inducing effect of PB or CBZ, and that neither protein binding nor erythrocyte distribution of ZNS could be the reason for the increased $Vd/F$ value of ZNS by VPA coadministration.

Keywords — zonisamide; pharmacokinetic interaction; antiepileptic drug; protein binding; erythrocyte distribution; metabolism

Introduction

Generally, antiepileptic drugs have a narrow margin of safety between effective and toxic levels. Furthermore, long-term medication and/or multi-drug therapy cannot be avoided because of the characteristics of convulsive disorders.

In recent years, zonisamide (1,2-benzoisoxazole-3-methanesulfonamide, ZNS), has been developed and used clinically for the treatment of various types of epilepsy.1-4) However, the pharmacokinetic behaviors of ZNS still remain unclear. In addition, the pharmacokinetic interaction of ZNS has not yet been fully elucidated,1,5) whereas a number of investigations have been reported for other antiepileptic drugs.6,7)

The present study was carried out to investigate the pharmacokinetics of ZNS and the effects of phenobarbital (PB), valproic acid (VPA), carbamazepine (CBZ) and phenytoin (PHT) on ZNS kinetics in rats. In addition, the effects of the drugs on the serum protein binding, erythrocyte distribution and metabolism of ZNS were also examined in vitro.

Materials and Methods

Materials — Zonisamide (ZNS) and N,N-dimethyl-ZNS were kindly supplied by Dainippon Pharmaceutical Co., Ltd (Tokyo, Japan). Phenobarbital (PB), sodium valproate (VPA-Na), carbamazepine (CBZ), phenytoin (PHT) and phenytoin sodium salt (PHT-Na) were obtained commercially. Acetonitrile was of high-performance liquid chromatography (HPLC) grade. All the other reagents were of analytical grade and were used without further purification.

Animal Study — Male Wistar rats weighing between 230 and 280 g were used and fasted for about 20 h before administration of the drugs.

ZNS Kinetics: In the intravenous experiments, ZNS dissolved in a physiological saline containing 40% polyethylene glycol 300 was ad-
ministered through a tail vein at a dose of 20 mg/kg. In the oral studies, rats received a single administration of ZNS suspended in 0.25% carboxymethylcellulose sodium salt (CMC-Na) solution at a dose of 10, 20, and 40 mg/kg, and received 20 mg/kg of ZNS at the same time of the day for 10 d as a multiple dosing.

Effects of Other Antiepileptics on ZNS Kinetics: The experiments were performed by two different dosage regimens as follows: (a) ZNS (20 mg/kg) suspended in 0.25% CMC-Na solution was coadministered orally with PB (20 mg/kg), VPA-Na (40 mg/kg as a free base) or CBZ (50 mg/kg); (b) after receiving oral administration of PB, VPA-Na or CBZ for 9 d as a pretreatment, ZNS was administered concomitantly with each drug. In the case of PHT, the experiments were made by intraperitoneal administration after dissolving PHT-Na (50 mg/kg as a free base) in a physiological saline containing 10% ethanol and 40% propylene glycol.

Blood samples were collected at designated time intervals from a jugular vein into heparinized tubes. Plasma obtained by centrifugation was kept at -20 °C until analysed.

In Vitro Study — Serum Protein Binding: Serum protein binding of ZNS was determined by an ultrafiltration technique at 25 °C using an Amicon Centrífree Micropartition System (Amicon Division, W.R. Grace & Co.-Conn., Beverly, MA, U.S.A.). ZNS was added to each serum obtained from normal rats to make a concentration of 10, 20, or 50 μg/ml. The serum protein binding of ZNS in the presence of other antiepileptics was determined by experiments performed at a concentration for each drug: 20 μg/ml for ZNS and 25 μg/ml for PB, 50 μg/ml for VPA, 10 μg/ml for CBZ, or 25 μg/ml for PHT.

Erythrocyte Distribution: Erythrocytes obtained from rat whole blood were washed three times with three volumes of 1/15 M phosphate buffer (pH 7.4, 300 mOsm), and finally suspended in the same solution. Erythrocyte suspensions were incubated with an equal volume of ZNS solution in a phosphate buffer at a final concentration of 10—500 μM for 20 min at 37 °C. The erythrocyte distribution of ZNS in the presence of other antiepileptic drugs was investigated by experiments performed at a concentration for each drug: 80 μM for PB, 300 μM for VPA, 30 μM for CBZ, or 50 μM for PHT. After incubation, the mixture was centrifuged to obtain the supernatant. Hematocrit value of the mixture was also determined in advance. After the supernatant (20 μl) was mixed with 30 μl of ethanol and 20 μl of internal standard solution, a 20-μl aliquot of this solution was directly injected onto the HPLC column.

Since it is considered that the concentration in the supernatant is in protein free form, the erythrocyte distribution ratio (C_b/C_f) of ZNS was calculated according to the following equation:

$$\frac{C_b}{C_f} = \frac{C_{\text{total}} - (1 - Ht)\cdot C_f}{Ht\cdot C_f}$$

where, C_b is ZNS concentration in erythrocytes, C_f is ZNS concentration in the supernatant, C_{total} is initial ZNS concentration in the mixture, and Ht is hematocrit.

Metabolism: ZNS metabolism was examined by using naive and pretreated rat hepatic microsomes, respectively. Rats were sacrificed after 24-h washout periods following multiple dosing of PB, VPA, CBZ or PHT in the same manner as mentioned above. Hepatic microsomes were prepared according to the method reported by Omura and Sato. Microsomal protein concentrations were determined by the method of Lowry et al. The metabolic rate of ZNS was measured in a 1-ml assay mixture containing microsomes (2 mg of microsomal protein), 0.1 M phosphate buffer (pH 7.4), 50 μM EDTA, 6 mM MgCl₂ and 1 μM ZNS. The metabolic rate of ZNS in the presence of other antiepileptics was determined by using naive rat microsomes and concentrations of substrates added were as follows: 1 μM for ZNS and 1.5 μM for PB, 1.5 μM for VPA, 0.5 μM for CBZ, or 1.5 μM for PHT. After 5 min of preincubation at 37 °C, the reaction was initiated by adding 100 μl of a mixture containing 10 mM NADP, 100 mM glucose-6-phosphate (G-6-P) and 20 unit/ml of G-6-P dehydrogenase. The incubation was carried out for 20 min and stopped by the addition of 100 μl of 6 M NaOH since the metabolic rate of ZNS was
linear up to 30 min at the substrate concentration used. Then, 100 μl of 0.1 M HCl was added in order to neutralize the reaction admixture. The metabolic rate of ZNS was calculated from the initial and final concentrations of substrate.

**Analytical Method** — The concentration of ZNS was determined by using HPLC technique as described previously. In this study, blood samples, 0.1 M phosphate buffer (pH 7.4) and N,N-dimethyl-ZNS (internal substance) used were 25 μl, 500 μl and 20 μl of a 50-μg/ml solution, respectively. The mobile phase used was a 70:11:15 mixture of 1% acetic acid, isopropyl alcohol, and acetonitrile.

**Data Analysis** — Pharmacokinetic parameters were computed by use of a nonlinear least squares regression program MULTI1. The area under the plasma concentration curve (AUC) was calculated by the trapezoidal rule from time 0 to 24 h, and the extrapolated residual area from the final concentration point to infinity. The statistical significance of the difference between groups was determined by the t test. A p value of <0.05 was considered to be statistically significant.

**Results**

**Pharmacokinetics of ZNS after Intravenous and Oral Administration**

![Graph](image)

**Fig. 1.** Time Course of Plasma Concentration of ZNS after Intravenous Administration of ZNS at a Dose of 20 mg/kg in Rats

Each point represents the mean ± S.E.M. of 4 rats and the solid line is fitted curve.

Figure 1 shows the time course of ZNS in the plasma after a bolus injection at a dose of 20 mg/kg. ZNS concentration declined from the plasma in a monoexponential manner. The mean ± S.D. values of elimination half-life (t_{1/2}), distribution volume (Vd) and AUC obtained in the present study were 9.2 ± 0.7 h, 1.10 ± 0.03 l/kg and 245.6 ± 16.9 μg·h/ml, respectively.

The time profiles of plasma concentration after a single oral administration of ZNS at a dose of 10, 20 and 40 mg/kg are shown in Fig. 2 (A),

![Graph](image)

**Fig. 2.** Time Courses of Plasma Concentration of ZNS after Single (A) and Multiple (B) Oral Administration of ZNS in Rats

Each point represents the mean ± S.E.M. of 4 rats and the solid lines are fitted curves. Dose (mg/kg): 40; 20; 10.
and the pharmacokinetic parameters estimated are listed in Table I. ZNS disappeared monoexponentially from the plasma with half-lives of 8.7—9.3 h at each dose examined. The maximum plasma levels were observed 2—4 h after administration and increased proportionally with dose. The bioavailability of ZNS determined by the $AUC$ method was 82—88%.

Figure 2 (B) shows the time course of plasma level over a dosage interval on day 10 of multiple dosing with ZNS (20 mg/kg). No significant difference was observed in the pharmacokinetic parameters after chronic dosing (Table I).

**Effects of PB, VPA, CBZ and PHT on ZNS**

![Graphs](image_url)

**Table I. Pharmacokinetic Parameters** of ZNS after Single or Multiple Oral Administration of ZNS in Rats

<table>
<thead>
<tr>
<th>Dose (mg/kg)</th>
<th>Single</th>
<th>Multiple$^b$</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>10</td>
<td>20</td>
</tr>
<tr>
<td></td>
<td>$k_a$</td>
<td>$t_{1/2}$</td>
</tr>
<tr>
<td>(h$^{-1}$)</td>
<td>(h)</td>
<td>(l/kg)</td>
</tr>
<tr>
<td>(0.52)</td>
<td>(0.4)</td>
<td>(0.07)</td>
</tr>
<tr>
<td>(1.56)</td>
<td>(0.6)</td>
<td>(0.11)</td>
</tr>
<tr>
<td>(1.24)</td>
<td>(1.5)</td>
<td>(0.09)</td>
</tr>
<tr>
<td>(1.24)</td>
<td>(1.5)</td>
<td>(0.09)</td>
</tr>
</tbody>
</table>

$^a$ Each value represents the mean (±S.D.) of 4 rats. $^b$ Multiple: 20 mg/kg, for 9 d. $^c$ Calculated from the normalized $AUC$ values of ZNS after oral and intravenous administrations.
Pharmacokinetic Interaction of ZNS

Table II. Pharmacokinetic Parameters\(^a\) of ZNS after Oral Administration of ZNS (20 mg/kg) in Combination with PB (20 mg/kg), VPA (40 mg/kg) or CBZ (50 mg/kg) in Rats

<table>
<thead>
<tr>
<th></th>
<th>ZNS alone</th>
<th>With PB</th>
<th>With PB(^b)</th>
<th>With VPA</th>
<th>With VPA(^b)</th>
<th>With CBZ</th>
<th>With CBZ(^b)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(k_t) (h(^{-1}))</td>
<td>1.56</td>
<td>1.80</td>
<td>2.35</td>
<td>2.22</td>
<td>2.02</td>
<td>1.14</td>
<td>1.47</td>
</tr>
<tr>
<td>(t_{1/2}) (h)</td>
<td>(0.31)</td>
<td>(0.13)</td>
<td>(1.23)</td>
<td>(0.92)</td>
<td>(0.53)</td>
<td>(0.32)</td>
<td>(0.36)</td>
</tr>
<tr>
<td>(Vd/F) (l/kg)</td>
<td>1.24</td>
<td>1.28</td>
<td>1.32</td>
<td>1.40</td>
<td>1.46(^c)</td>
<td>1.19</td>
<td>1.32</td>
</tr>
<tr>
<td>(AUC) (μg·h/ml)</td>
<td>215.6</td>
<td>197.0</td>
<td>179.5</td>
<td>177.4(^a)</td>
<td>181.8(^d)</td>
<td>206.5</td>
<td>175.1(^e)</td>
</tr>
</tbody>
</table>

\(^a\) Each value represents the mean (±S.D.) of 4—6 rats. \(^b\) Pretreated by oral administration of PB (20 mg/kg), VPA (40 mg/kg) or CBZ (50 mg/kg) for 9 d. \(^c\) \(p<0.01\), vs. corresponding control. \(^d\) \(p<0.05\), vs. corresponding control. \(^e\) \(p<0.05\), vs. corresponding simultaneous administration group.

Kinetics

The time courses of plasma ZNS level after simultaneous oral administration of ZNS (20 mg/kg) with PB (20 mg/kg), VPA (40 mg/kg) or CBZ (50 mg/kg) in rats untreated and pretreated with each drug are illustrated in Fig. 3 (A—C), and the pharmacokinetic parameters are summarized in Table II when ZNS was administered alone (Table I).

The \(t_{1/2}\) and \(AUC\) values of ZNS in rats pretreated with PB were lower than those in other two groups, although no significant difference was observed. The \(k_t\), first-order absorption rate constant, and \(Vd/F\) values showed no significant difference in each group examined.

In the case of VPA, no significant difference in the \(t_{1/2}\) value of ZNS was observed among three groups studied. On the other hand, significantly high \(Vd/F\) and low \(AUC\) values of ZNS in rats pretreated with VPA were observed compared with those obtained by ZNS alone.

The \(t_{1/2}\) and \(AUC\) values of ZNS showed a significant decrease after multiple dosing of CBZ, whereas no significant change in the pharmacokinetic parameters was observed in the simultaneous administration group.

It has been reported that the absorption of PHT varied widely because of its poor solubility.\(^{12}\) In the present study, thus, the drugs were

![Fig. 4. Time Courses of Plasma Concentration of ZNS after Intraperitoneal Administration of ZNS (20 mg/kg) Alone and in Combination with PHT (50 mg/kg) in Rats](image)

Each point represents the mean ±S.E.M. of 4—6 rats and the solid lines are fitted curves. ○, untreated; □, pretreated by intraperitoneal administration of PHT (50 mg/kg) for 9 d; ▲, ZNS alone.

Table III. Pharmacokinetic Parameters\(^a\) of ZNS after Intraperitoneal Administration of ZNS (20 mg/kg) in Combination with PHT (50 mg/kg) in Rats

<table>
<thead>
<tr>
<th></th>
<th>ZNS alone</th>
<th>With PHT</th>
<th>With PHT(^b)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(k_t) (h(^{-1}))</td>
<td>12.09</td>
<td>26.10</td>
<td>20.25</td>
</tr>
<tr>
<td>(t_{1/2}) (h)</td>
<td>(2.30)</td>
<td>(13.92)</td>
<td>(9.92)</td>
</tr>
<tr>
<td>(Vd/F) (l/kg)</td>
<td>8.2</td>
<td>8.3</td>
<td>8.1</td>
</tr>
<tr>
<td>(AUC) (μg·h/ml)</td>
<td>(0.04)</td>
<td>(0.09)</td>
<td>(0.06)</td>
</tr>
</tbody>
</table>

\(^a\) Each value represents the mean (±S.D.) of 4—6 rats. \(^b\) Pretreated by intraperitoneal administration of PHT (50 mg/kg) for 9 d.
administered intraperitoneally to grasp accurately the pharmacokinetic interaction between ZNS and PHT. Figure 4 shows the time profiles of plasma concentration of ZNS following intraperitoneal coadministration of ZNS (20 mg/kg) with PHT (50 mg/kg) in rats untreated and pretreated with PHT and those obtained with ZNS alone. There showed no significant change in the pharmacokinetic parameters of

ZNS obtained by each experiment (Table III).

In Vitro Serum Protein Binding of ZNS

The bound fractions of ZNS were 28.6—33.3%, and showing no significant difference among the three concentrations of ZNS (10, 20 and 50 μg/ml).

The effects of other antiepileptics on the serum protein binding of ZNS are shown in Table IV. No significant difference in the bound fraction of ZNS was observed in the presence of PB, VPA, CBZ or PHT.

In Vitro Erythrocyte Distribution of ZNS

The erythrocyte distribution of ZNS was studied and the $C_b/C_v$ value (26.8—2.7) decreased nonlinearly with increasing ZNS concentration. Then, the binding parameters of ZNS to erythrocytes are calculated by use of Scatchard plots as shown in Fig. 5 and are listed in Table V. There showed two components: high- and low-affinity components.

The effects of other antiepileptic drugs on the erythrocyte distribution of ZNS are summarized in Table V. No marked change was observed in the binding parameters of ZNS compared with those of corresponding control.

In Vitro Metabolism of ZNS

Table VI shows the effect of PB, VPA, CBZ or PHT pretreatment on the metabolic rate of

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**Table IV. Serum Protein Binding of ZNS (20 μg/ml) in the Presence of Other Antiepileptics**

<table>
<thead>
<tr>
<th>Binding ratio** of ZNS (%)</th>
<th>Without PB</th>
<th>With PB</th>
<th>Without VPA</th>
<th>With VPA</th>
<th>Without CBZ</th>
<th>With CBZ</th>
<th>Without PHT</th>
<th>With PHT</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>30.4</td>
<td>28.6</td>
<td>31.9</td>
<td>32.0</td>
<td>28.6</td>
<td>31.2</td>
<td>31.2</td>
<td>31.3</td>
</tr>
<tr>
<td></td>
<td>(4.2)</td>
<td>(2.5)</td>
<td>(2.1)</td>
<td>(3.7)</td>
<td>(2.5)</td>
<td>(1.5)</td>
<td>(4.2)</td>
<td>(4.2)</td>
</tr>
</tbody>
</table>

*a) PB, 25 μg/ml; VPA, 50 μg/ml; CBZ, 10 μg/ml; PHT, 25 μg/ml. b) Each value represents the mean (± S.D.) of 3—5 rats.

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**Table V. Binding Parameters**

<table>
<thead>
<tr>
<th>Binding Parameters** of ZNS to Erythrocytes in the Presence of Other Antiepileptics</th>
<th>ZNS alone</th>
<th>With PB</th>
<th>With VPA</th>
<th>With CBZ</th>
<th>With PHT</th>
</tr>
</thead>
<tbody>
<tr>
<td>$K_{A1}$ (μM$^{-1}$)</td>
<td>0.173</td>
<td>0.167</td>
<td>0.181</td>
<td>0.193</td>
<td>0.169</td>
</tr>
<tr>
<td>$B_{max1}$ (μM)</td>
<td>168.1</td>
<td>161.2</td>
<td>163.7</td>
<td>162.3</td>
<td>167.9</td>
</tr>
<tr>
<td>$B_{A_{RBC1}}$ (μM)</td>
<td>29.1</td>
<td>26.9</td>
<td>29.6</td>
<td>31.3</td>
<td>28.4</td>
</tr>
<tr>
<td>$K_{a2}$ (μM$^{-1}$)</td>
<td>0.0013</td>
<td>0.0018</td>
<td>0.0014</td>
<td>0.0017</td>
<td>0.0011</td>
</tr>
<tr>
<td>$B_{max2}$ (μM)</td>
<td>2817.7</td>
<td>2081.9</td>
<td>2733.4</td>
<td>2374.6</td>
<td>3541.2</td>
</tr>
<tr>
<td>$B_{A_{RBC2}}$ (μM)</td>
<td>3.7</td>
<td>3.8</td>
<td>3.8</td>
<td>4.0</td>
<td>3.9</td>
</tr>
</tbody>
</table>

*a) Subscripts 1 and 2 denote for high and low affinity, respectively. b) PB, 80 μM; VPA, 300 μM; CBZ, 30 μM; PHT, 50 μM. c) Association constant. d) Binding capacity. e) Binding ability ($=K_{A1}B_{max}$).
Pharmacokinetic Interaction of ZNS

### Table VI. Inducing Effect of Other Antiepileptics on Metabolism of ZNS (1.0 μM) by Liver Microsomes in Rats

<table>
<thead>
<tr>
<th></th>
<th>Control</th>
<th>PB&lt;sup&gt;b&lt;/sup&gt;</th>
<th>VPA&lt;sup&gt;b&lt;/sup&gt;</th>
<th>CBZ&lt;sup&gt;b&lt;/sup&gt;</th>
<th>Control</th>
<th>PHT&lt;sup&gt;c&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Metabolic rate&lt;sup&gt;a&lt;/sup&gt; of ZNS (pmol/min/mg protein)</td>
<td>5.91</td>
<td>9.15&lt;sup&gt;d&lt;/sup&gt;</td>
<td>5.58</td>
<td>14.49&lt;sup&gt;d&lt;/sup&gt;</td>
<td>6.25</td>
<td>6.03</td>
</tr>
<tr>
<td></td>
<td>(0.77)</td>
<td>(0.92)</td>
<td>(1.27)</td>
<td>(2.07)</td>
<td>(0.79)</td>
<td>(0.89)</td>
</tr>
</tbody>
</table>

<sup>a</sup> Each value represents the mean (± S.D.) of 3 rats. <sup>b</sup>Pretreated by oral administration of PB (20 mg/kg), VPA (40 mg/kg) or CBZ (50 mg/kg) for 9 d. <sup>c</sup>Pretreated by intraperitoneal administration of PHT (50 mg/kg) for 9 d. <sup>d</sup>p < 0.01, vs. corresponding control.

### Table VII. Effect of Other Antiepileptics<sup>d</sup> on Metabolism of ZNS (1.0 μM) by Liver Microsomes in Rats

<table>
<thead>
<tr>
<th></th>
<th>ZNS alone</th>
<th>With PB</th>
<th>With VPA</th>
<th>With CBZ</th>
<th>With PHT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Metabolic rate&lt;sup&gt;b&lt;/sup&gt; of ZNS (pmol/min/mg protein)</td>
<td>5.68</td>
<td>5.44</td>
<td>5.28</td>
<td>5.42</td>
<td>4.54</td>
</tr>
<tr>
<td></td>
<td>(0.98)</td>
<td>(0.98)</td>
<td>(0.29)</td>
<td>(0.79)</td>
<td>(1.86)</td>
</tr>
</tbody>
</table>

<sup>a</sup> PB, 1.5 μM; VPA, 1.5 μM; CBZ, 0.5 μM; PHT, 1.5 μM. <sup>b</sup> Each value represents the mean (± S.D.) of 3 rats.

ZNS. There showed a significant increase of metabolic rate of ZNS following PB and CBZ pretreatment, whereas no significant change in the metabolic rate of ZNS after multiple dosing of VPA and PHT.

The metabolic rates of ZNS in the presence of PB, VPA, CBZ or PHT are listed in Table VII. No significant change in the metabolic rate of ZNS was observed in all cases.

### Discussion

In the present study, we investigated the pharmacokinetics of ZNS after single or multiple administration, and found that ZNS declined monoexponentially from the plasma and showed a linear disposition kinetics. Moreover, the pharmacokinetics of ZNS were not changed after multiple dosing. These findings were in good agreement with those reported by Matsumoto et al.<sup>13,14</sup>

The bound fractions of ZNS in rats ranged from 28.6 to 33.3%, which were lower than those obtained in our previous study in human (38.3—39.8%).<sup>10</sup> In addition, there showed a nonsaturating binding phenomenon within the range examined as well as in human. The $C_p/C_t$ values were 2.7—26.8, indicating that ZNS is significantly concentrated into erythrocytes. ZNS binding to erythrocytes showed high- and low-affinity components, respectively. As reported by Matsumoto et al.,<sup>15,16</sup> it seems that the former is corresponding to binding of ZNS to carbonic anhydrase and the latter is to other erythrocyte protein.

It is well known that the serum protein binding, erythrocyte distribution and metabolism are the possible factors affecting the drug kinetics.<sup>17—20</sup> Therefore, the effect of other antiepileptics on these factors of ZNS were investigated to ascertain the mechanism of the drug interactions in vitro.

It has been reported that PB, CBZ and PHT increase the liver microsomal enzymes responsible for the drug metabolism, resulting a decreased plasma level of a concurrent drug.<sup>6,7,21,22</sup> Furthermore, Konishi et al.<sup>23</sup> investigated the enzyme inducing effects of PB and CBZ in mouse hepatic microsomes and found the enhanced oxidative enzyme activity. In the present study, the metabolic rate of ZNS increased after multiple dosing of PB or CBZ, indicating the enhanced metabolism of ZNS. On the contrary, there showed no significant alteration in the serum protein binding and erythrocyte distribution of ZNS. These results suggest that the decreased $t_{1/2}$ value of ZNS might be attributable to the inducing effect of PB or CBZ.
The pharmacokinetic behaviors and the metabolic rate of ZNS were not influenced by PHT pretreatment. Ojemann et al., however, reported the decreased $t_{1/2}$ of ZNS in patients treated with PHT monotherapy. It seems that further detailed study should be made to clarify the mechanism of ZNS-PHT interaction.

It is known that VPA inhibits the drug-metabolizing enzymes and delays the elimination of PB. In the present study, however, no significant difference in the $t_{1/2}$ value of ZNS was observed among three group examined. In addition, no marked change in the metabolic rate of ZNS was observed in the presence of VPA. These results indicate that VPA has no effect on the metabolism of ZNS. On the contrary, there showed a significant increase in the $Vd/F$ value of ZNS after multiple administration of VPA. It has been also reported that VPA displaces a drug from its plasma protein binding sites, resulting in an increased drug distribution to extravascular tissue. However, no significant change by VPA was observed in the serum protein binding and erythrocyte distribution of ZNS. As a result, it is concluded that neither protein binding nor erythrocyte distribution of ZNS could be the reason for the increased $Vd/F$ value of ZNS by VPA coadministration. Although the $Vd$ and $F$ values were not separately evaluated in this study, it might in part be explained by the decreased $F$ value. It seems, however, a more detailed study should be made.

In the present experiments, we investigated the pharmacokinetics of ZNS and the pharmacokinetic interaction between ZNS and PB, VPA, CBZ or PHT in rats. The results demonstrated the following: (1) ZNS showed a linear disposition kinetics within the doses examined; (2) marked decrease in the $t_{1/2}$ value of ZNS was observed by the enzyme inducing effect of PB or CBZ; (3) there showed the increased $Vd/F$ value of ZNS by VPA pretreatment; (4) there was no marked effect of PHT on ZNS kinetics; (5) there was little effect of other antiepileptic drugs on the serum protein binding and erythrocyte distribution of ZNS in vitro.

References


Pharmacokinetic Interaction of ZNS


