J. Pharm. Pharmacol. 1996, 48: 1270–1275
Received March 5, 1996
Accepted March 21, 1996

Pharmacokinetic Study of Zonisamide in Patients Undergoing Brain Surgery

ICHIRO IEIRI, TAKATO MORIOKA*, SONYORI KIM, SHUNJI NISHIO*, MASASHI FUKUI* AND SHUN HIGUCHI

*Division of Pharmaceutical Science, Kyushu University and *Department of Neurosurgery Kyushu University Hospital, Fukuoka, Japan*

Abstract

To test whether the concentration of the anticonvulsant zonisamide in erythrocytes reflects the brain concentration and the clinical response of the drug, its pharmacokinetics were studied in nine patients undergoing surgery for brain tumour. Erythrocyte, total, and free serum concentrations in samples drawn on the day of brain surgery were compared with levels on a day after the operation. In three patients zonisamide and its major metabolite, 2-sulphamoylacetylphenol, were also analysed in urine. The area under the curve of the free and the erythrocyte concentration did not differ between the two study phases whereas the area under the curve of the total serum concentration was significantly lower on the day of the operation, and this was associated with significant increases in total clearance (15·4 compared with 12·7 mL kg^{-1} h^{-1}, $P < 0.05$, $n = 9$) and renal clearance (5·4 compared with 3·3 mL kg^{-1} h^{-1}, $P < 0.05$, $n = 3$), and non-significant change in non-renal clearance (7·7 on the day of operation compared with 8·4 mL kg^{-1} h^{-1} on the post-operation day, $n = 3$). Zonisamide distribution was also altered by the operative procedure, as evidenced by a higher volume of distribution (1·48 compared with 0·87 L kg^{-1}, $P < 0.05$, $n = 9$). The binding of zonisamide was characterized on both days. Zonisamide binding to erythrocytes seemed to occur by two processes: a saturable process and a non-saturable linear process. The maximum binding capacity to erythrocytes (31·6 vs 29·7 μg mL^{-1}) did not differ on the two days; however, increases in the dissociation binding constant (+28%) and the proportionality constant (+24%) were observed on the day of the operation, suggesting that the zonisamide concentration in erythrocytes was greater on the day of the operation.

Brain surgery appears to be one of the possible factors altering the rate of elimination of zonisamide and the uptake of the drug by erythrocytes.

The prophylactic administration of antiepileptic drugs (AEDs) to patients undergoing brain surgery is a standard practice. Characterization of the pharmacokinetics of AEDs during surgical procedures is therefore necessary for their clinical use. We have previously reported the pharmacokinetic properties of valproic acid during brain surgery, and showed that there was an alteration in valproic acid disposition on the day of the operation (Ieiri et al 1995). No information is available for other AEDs in this situation, however. In recent years, zonisamide (3-sulphamoylmethyl-1,2-benzisoxazole) has been developed and used clinically for the treatment of various types of epilepsy in Japan (Sackellares et at 1985; Wilensky et al 1985). The disposition kinetics of zonisamide differ from those of valproic acid. Zonisamide undergoes some metabolism involving glucuronic acid conjugation, acetylation, and hydroxylation: in a preliminary study in man, unchanged zonisamide (30–50% of the dose) and glucuronide of 2-sulphamoylacetylphenol (10–20% of the dose) were detected with a minor component of acetylated zonisamide in urine, and only the unchanged zonisamide was detected in plasma (Ito et al 1982; Matsumoto et al 1983). Zonisamide has low plasma protein binding to albumin, of the order of about 50% (Peters & Sorkin 1993) and it is highly bound to erythrocytes within the therapeutic range (Matsumoto et al 1989a). The drug concentration in erythrocytes has been claimed to reflect the brain concentration and the clinical response of the drug (Garver et al 1977; Casper et al 1980). This study was designed to investigate the effects of brain surgery on the pharmacokinetic properties of prophylactic zonisamide.

Correspondence: I. Ieiri, Division of Pharmaceutical Science, Kyushu University, Maidashi 3-1-1, Higashi-ku, Fukuoka, 812, Japan.

Materials and Methods

Patients

The pharmacokinetics of zonisamide were evaluated in nine adults undergoing surgery for brain tumours. Individual patient variables are listed in Table 1. All patients received zonisamide (Exceglan; Dainippon Pharmaceutical, Japan), given in the morning for at least 2 weeks before the operation as prophylaxis against postoperative epilepsy. None of the patients had taken other antiepileptic drug(s) which could have interfered with zonisamide kinetics, and none showed any evidence of liver or renal dysfunction during the study period. Written informed consent was obtained from all patients, and the protocol of the study was approved by the local ethics committee.

Sampling

On the day of the operation, blood samples were taken at 08:00 h, just before drug administration, and at 2-h intervals for the first 4 h then collection at 8, 10, 14 and 24 h after drug administration. On the 4th to 8th day after the operation, when the patients had recovered from surgery, blood samples (approximately 3·5 mL) were taken at 09:00 h, just before drug administration, and thereafter 2, 4, 8, 12, and 24 h after drug administration. The sampling time protocol was modified slightly according to the patient's clinical situation. In three patients, urine samples were collected for 24 h after drug

Table 1. Details of patients.

	Patient number:								
	1	2	3	4	5	6	7	8	9
Sex	M	M	M	F	F	F	M	F	F
Age (years)	30	50	69	60	40	50	51	37	53
Zonisamide dose (mg kg^{-1} day^{-1})	2·3	3·5	3·3	3·4	2·2	1·9	3·3	3·6	3·7
Length of operation (h)	8·3	6·6	3·9	4·8	5·5	4·7	6·5	6·0	9·8
Blood loss (g)	500	200	600	600	910	500	600	550	800
Urine volume (mL)	1900	1800	1200	900	1300	1400	400	700	2200
Volume of intravenous fluid (mL)	3800	3200	3000	2350	2450	2900	1300	2900	4250
Interval between day of operation and post operation day	8	7	6	4	4	4	7	4	6

administration and the volume was recorded on both days. Serum and urine samples were stored at − 20°C and erythrocytes were stored at 4°C until assayed.

Treatment

All surgical procedures were started at approximately 10:00 h. Anaesthesia was induced in all patients with oxygen and nitrous oxide in combination with fentanyl (11·8 ± 7·0 μg kg^{-1}, mean ± s.d.) and droperidol (0·14 ± 0·04 mg kg^{-1}). Muscle paralysis was induced with vecuronium (0·18 ± 0·06 mg kg^{-1}). Atropine (0·5 mg) and nitrazepam (5·5 ± 0·8 mg) were given as premedication on the night before the day of the operation. During the operation all patients received 1·3 to 4·3 L of intravenous fluid containing glucose and electrolytes. None received concentrated red blood cells.

Measurement of drug concentration

The concentrations of zonisamide and 2-sulphamoylacetylphenol in samples were measured in duplicate by high performance liquid chromatography (HPLC). Venous blood was collected in a plain syringe and the serum was separated at room temperature. The serum (1 mL) was centrifuged for 10 min at 3000 rev min^{-1} (10°C), using an ultrafiltration technique (Amicon Micropartition MPS-3 System; Amicon, Danvers, MA, USA) to obtain unbound zonisamide.

For HPLC analysis of total and free serum zonisamide concentrations, phosphate buffer (100 mM, pH 6·8; 1·0 mL), containing allobarbital as internal standard, was added to the sample (0·1 mL). The mixture (1 mL) was poured into an Extrelut-1 column (E. Merck). After 10 min, the column was eluted with *tert*-butyl methyl ether (3·0 mL). The eluate was dried and then dissolved in methanol (100 μL); 20-μL volumes were injected into the chromatograph consisting of a Shimadzu (Kyoto, Japan) LC-10A system equipped with a LC-10AS pump and an SPD-10A UV detector. The column (75 mm × 4 mm i.d.) was a LiChro-Cart HPLC Cartridge Superspher RP-18, 4-μm particle diameter (E. Merck). The column temperature was ambient. The mobile phase was 20% methanol. The flow rate was 1·2 mL min^{-1}, and the eluates were monitored at 210 nm.

For HPLC analysis of urine, phosphate buffer (100 mM, pH 5·0; 1·0 mL), containing allobarbital as internal standard, was added to urine (0·1 mL). Because the amount of free 2-sulphamoylacetylphenol in the urine sample was too low to be detected, the hydrolysis of the 2-sulphamoylacetylphenol conjugate was performed before the assay. 2-Sulphamoylacetylphenol glucuronide in the urine sample was treated with 400 units of β-glucronidase at 37°C for 18 h, after which the mixture (1·0 mL) was poured into an Extrelut-1 column.

Fresh heparinized whole blood was centrifuged, and the plasma removed. For HPLC analysis of erythrocyte zonisamide concentrations, distilled water (0·15 mL) and methanol (0·95 mL), containing allobarbital as internal standard, were added to erythrocytes (0·2 mL). The resulting mixture was shaken for 10 min and centrifuged at 3500 rev min^{-1} for 5 min. The supernatant (0·1 mL) was mixed with phosphate buffer (100 mM, pH 6·8; 1·0 mL), and 1·0 mL of the mixture was poured into an Extrelut-1 column.

The sensitivity of the zonisamide assay in samples was 50 ng mL^{-1}; the intra- and inter-assay coefficients of variation were 1·9 and 1·4%, respectively. The respective values for the 2-sulphamoylacetylphenol assay in urine were 50 ng mL^{-1}, 2·5%, and 0·3%.

Pharmacokinetic analysis

Zonisamide pharmacokinetic parameters were estimated by appropriate compartment modelling and by model-independent methods. The areas under the zonisamide concentration-time curves (AUC) were calculated by the trapezoidal rule from 0 to 24 h after drug administration. Steady-state conditions were confirmed by comparison of the concentrations 0 and 24 h post-administration: a non-significant difference between these two levels was taken as an evidence that a steady state had been achieved. Total clearance (CL_t) was calculated according to the equation $CL_t = \text{dose}/AUC_s$, where AUC_s is the AUC of the total serum concentration and dose is the daily dose (mg kg^{-1}). Zonisamide renal clearance (CL_r) was determined by dividing the amount of zonisamide excreted into the urine during a collection interval by the AUC_s for that interval. Because the formation of 2-sulphamoylacetylphenol is mediated by P4503A (Nakasa et al 1993) and isoxazole cleavage of zonisamide probably occurs in the liver and also the gut wall, non-renal clearance (CL_{nr}) was estimated from the equation $CL_{nr} = CL_t - CL_r$. The elimination rate constant (k_e) was estimated by least squares regression analysis from the terminal post-distributive phase of the serum concentration-time curve, after which the apparent volume of distribution (Vd) was calculated from $Vd = CL_t/k_e$.

The binding parameters for zonisamide to erythrocytes were estimated by non-linear least squares regression from the equation $C_e = B_{max} \times C_s/(k_d + C_s) + k \times C_s$, where C_e is the erythrocyte concentration of zonisamide, B_{max} is the maximum

binding capacity ($\mu g\ mL^{-1}$), C_s is the serum concentration of zonisamide, k_d is the dissociation binding constant ($\mu g\ mL^{-1}$) and k is the proportionality constant relating to the serum concentration of zonisamide (Boddy et al 1989).

Statistical analysis

Statistical comparisons were made with Student's paired *t*-test. All values were expressed as means ± standard deviation (s.d.), and probability values of < 5% were considered significant.

Results

The mean ± s.d. zonisamide concentration–time profiles on the day of the operation and on the post-operation day are shown graphically in Fig. 1. Although the free zonisamide concentrations in serum (C_f) and erythrocytes concentration (C_e) did not differ between these two study phases, the mean total zonisamide concentration in serum (C_s) on the day of the operation was lower than that on the post-operation day throughout the observation period. The pharmacokinetic parameters are summarized in Table 2. The mean AUC_f and AUC_e were not significantly different between the day of the operation and the post-operation day. In contrast, the mean AUC_s was significantly lower on the day of the operation, associated with significant increases in CL_t and Vd.

Urinary excretion of zonisamide and 2-sulphamoylacetylphenol could be measured for three of the nine patients only; the results are shown in Table 3. The mean urinary recovery, over 24 h, of zonisamide was significantly higher (40·5% of the administered dose) on the day of the operation than on the post-operation day (25·9%). The mean urinary excretion of 2-sulphamoylacetylphenol tended also to be higher on the day of the operation. The mean renal clearance of zonisamide ($5{\cdot}4\ mL\ kg^{-1}\ h^{-1}$) on the day of the operation was significantly higher than that ($3{\cdot}3\ mL\ kg^{-1}\ h^{-1}$) on the post-operation day. In contrast, the non-renal clearance did not differ between the two study phases. Renal elimination of zonisamide was considered to be faster on the day of the operation, as indicated by the greater renal clearance and the greater fraction excreted unchanged in the urine over the collection period compared with those parameters obtained on the post-operation day.

The plots of zonisamide concentration in erythrocytes against that in serum are shown in Fig. 2; the binding parameters for zonisamide to erythrocytes are given in the inset of the figure. Zonisamide was markedly concentrated in erythrocytes, with both saturable and non-saturable components, as previously described by Matsumoto et al (1989b). The B_{max} on the day of the operation was comparable with that on the post-operation day, but the estimated k and k_d values on the day of the operation were higher than the postoperative values.

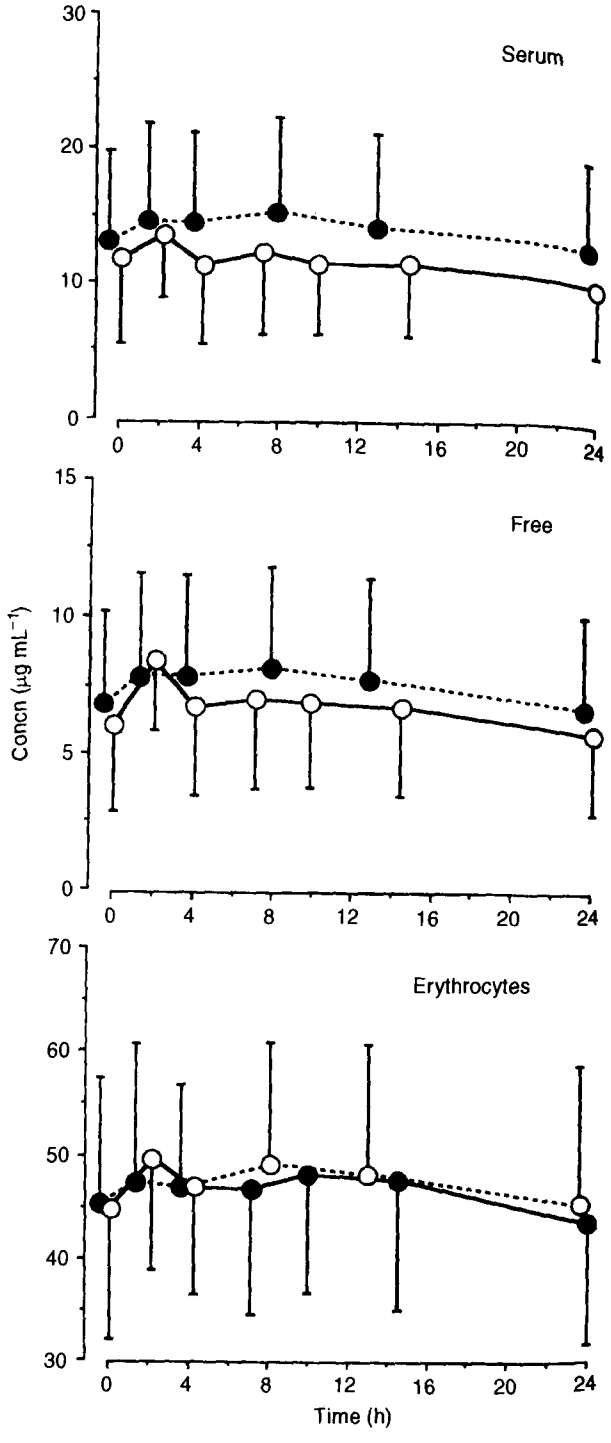

FIG. 1. Mean zonisamide concentration–time profiles on the day of the operation (○) and the post-operation day (●).

Discussion

In this study, there were no significant differences between values of AUC_f and AUC_e on the day of the operation and those the post-operation day. AUC_s was, on the other hand, significantly lower on the day of the operation than on the post-operation day. The CL_t values obtained from the samples on both days were similar to those reported for epileptic patients not undergoing surgery (Wilensky 1984; Ojemann et al 1986), but the mean CL_t on the day of the operation was significantly higher than the mean post-operation value.

As demonstrated, the mean renal elimination of zonisamide was 1·6 times faster on the day of the operation than on the post-operation day. It is well known that the urine flow rate is higher in the hydrated condition than in the non-hydrated condition, and this can affect the renal excretion of various

Table 2. Pharmacokinetic parameters on the day of the operation and the post-operation day.

		Patient number: 1	2	3	4	5	6	7	8	9	Mean ± s.d.
Area under the total serum concentration–time curve ($\mu g\,h\,mL^{-1}$)	Day of operation	329·1	236·0	228·3	338·7	114·2	41·2	345·6	475·6	353·4	273·6* ± 133·5
	Post-operation day	353·0	304·2	428·6	384·2	121·2	47·7	404·7	588·0	410·6	338·0 ± 164·0
Area under the free concentration–time curve ($\mu g\,h\,mL^{-1}$)	Day of operation	211·0	165·1	141·6	208·8	64·6	18·4	180·1	265·6	205·3	162·3 ± 77·5
	Post-operation day	197·5	204·7	233·4	235·5	61·1	22·7	203·0	280·4	230·0	185·4 ± 85·6
Area under the erythrocyte concentration–time curve ($\mu g\,h\,mL^{-1}$)	Day of operation	-†	1158·7	1046·1	1110·7	934·0	714·3	856·3	1652·4	1394·5	1108·4 ± 300·8
	Post-operation day	-†	1074·8	1223·6	1102·8	902·3	736·9	944·4	1637·3	1416·4	1134·3 ± 300·1
Total clearance‡ ($mL\,kg^{-1}\,h^{-1}$)	Day of operation	6·94	14·61	14·60	10·09	19·46	45·00	9·41	7·51	10·58	15·44* ± 11·75
	Post-operation day	6·48	11·34	7·78	8·90	18·34	38·86	8·04	6·07	9·11	12·67 ± 10·51
Apparent volume of distribution‡ ($L\,kg^{-1}$)	Day of operation	1·15	2·26	3·42	1·07	0·82	1·16	0·81	1·38	1·27	1·48* ± 0·84
	Post-operation day	0·89	0·79	0·63	0·87	1·10	0·85	0·83	1·06	0·78	0·87 ± 0·15
Ratio of the area under the free concentration–time curve to that under the total serum concentration–time curve	Day of operation	0·64	0·70	0·62	0·62	0·57	0·45	0·52	0·56	0·58	0·58* ± 0·07
	Post-operation day	0·56	0·67	0·54	0·61	0·50	0·48	0·50	0·48	0·56	0·54 ± 0·06

*$P < 0·05$ compared with the data on the post-operation day. †No data. ‡Calculated from the total serum concentration.

Table 3. Urinary excretion and estimated renal and non-renal clearance on day of operation and post-operation day.

		Patient number: 5	7	9	Mean ± s.d.
Urinary excretion of zonisamide as percentage of the dose administered	Day of operation	41·1	38·9	41·6	40·5* ± 1·4
	Post-operation day	32·8	19·9	25·2	25·9 ± 6·5
Urinary excretion of 2-sulphamoyl-acetylphenol as percentage of the dose administered	Day of operation	5·3	5·7	12·9	8·0 ± 4·3
	Post-operation day	1·8	4·9	2·5	3·1 ± 1·6
Renal clearance ($mL\,kg^{-1}\,h^{-1}$)	Day of operation	8·0	3·7	5·4	5·4* ± 2·3
	Post-operation day	6·0	1·6	2·3	3·3 ± 2·4
Non-renal clearance ($mL\,kg^{-1}\,h^{-1}$)	Day of operation	11·0	5·3	6·6	7·7 ± 3·0
	Post-operation day	12·0	6·4	6·7	8·4 ± 3·2

*$P < 0·05$ compared with the data on post-operation day.

drugs and other substances (Faruq et al 1990; Voltonen et al 1993); there is, nevertheless, no simple explanation for the increased renal excretion of zonisamide on the day of the operation, because the fundamental mechanisms of excretion are not well understood. Because zonisamide is partially excreted by the kidneys (30–50% of the administered dose), it is conceivable that the increased urinary flow rate resulting from the administration of intravenous fluid during surgery might account for the partial increase. Indeed, the urine flow-rate on the day of the operation was significantly higher than that on the post-operation day in the three patients for whom we obtained this measurement ($2·7 \pm 0·7$ L day^{-1} compared with $1·4 \pm 0·4$ L day^{-1}, $P < 0·01$). It remains unclear, however, why the excretion of 2-sulphamoylacetylphenol in this situation was also high. In contrast with the CL_r, the estimated non-renal clearance including hepatic clearance, did not differ between the two study days. The lack of a significant effect of brain surgery on zonisamide non-renal clearance was our most surprising result. Although the number of patients studied is too small to reach a definite conclusion, our findings suggest that the increase in CL_t on the day of the operation might, in part, have been a result of the increased CL_r.

Sulphonamides such as zonisamide with aromatic substituents have a high affinity for carbonic anhydrase and are

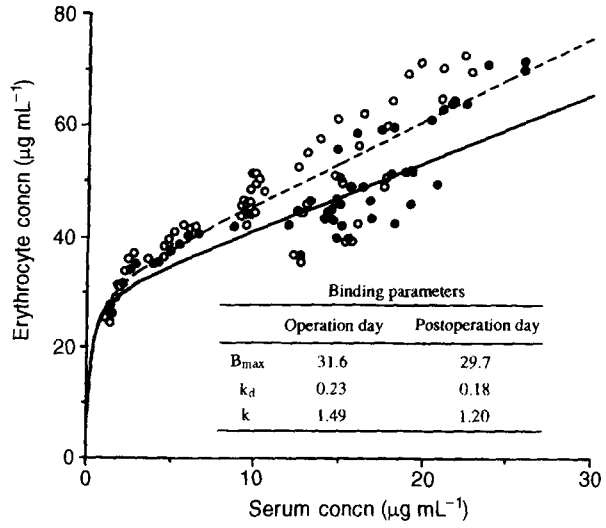

FIG. 2. Relationship between serum and erythrocyte concentrations of zonisamide, and the binding parameters to erythrocytes on the day of the operation (○) and the post-operation day (●). B_{max}, k_d and k indicate the maximum binding capacity (μg mL^{-1}), dissociation binding constant (μg mL^{-1}), and proportionality constant, respectively, for the serum concentration.

thereby concentrated in erythrocytes to a significant extent, because these cells contain a large amount of the enzyme (Maren 1976; Funakoshi & Deutsch 1968). Matsumoto et al (1989a) reported two uptake processes for zonisamide, saturable binding to carbonic anhydrase and non-saturable distribution, and they revealed that not only carbonic anhydrase but also other erythrocyte proteins were responsible for the concentration of sulphonamides in human erythrocytes. The saturable, high-affinity component corresponds to the binding of zonisamide to carbonic anhydrase and the non-saturable, low-affinity component corresponds to the binding of zonisamide to other proteins.

In this study, the dissociation binding constant and the proportionality constant were higher on the day of the operation, indicating that the zonisamide was concentrated in erythrocytes to a greater extent on this day than on the day after. Several factors during surgery, such as anaesthetic agents, volume repletion, and fluid redistribution, secondary to surgical insults might influence erythrocyte binding. It is, however, difficult to implicate anaesthesia as a cause of the increase in kd and k, and, to the best of our knowledge, there have been no reports suggesting that general anaesthesia has an effect on the binding of zonisamide to erythrocytes. A possible explanation of the observed high kd and k is changes in serum albumin level as a result of blood loss and haemodilution. It has been shown that the concentration of zonisamide in erythrocytes is affected by the serum albumin level (Driessen et al 1989) and several other reports (Evans & Shand 1973; Kurata & Wilkinson 1974; Hughes et al 1976) have indicated that extensive plasma protein binding reduces the uptake of the drug by red blood cells by reducing the free plasma drug concentration. Matsumoto et al (1989b) also found that the concentration of zonisamide in erythrocytes was affected by the presence of extracellular albumin. Their in-vitro study indicated that the concentration of zonisamide in erythrocytes was higher under low-albumin conditions, and that this was associated with an increase in the proportionality constant. In our study, because the serum albumin level on the day of the operation was significantly lower than that on the post-operation day, probably because of intra-operative blood loss and haemodilution, the intra-operative free fraction of zonisamide was significantly higher than the post-operative value. This increase in the free fraction brought about by surgery was also seen with valproic acid in a similar group of patients (Ieiri et al 1995). In this study a significantly positive relationship was observed between the free fraction of zonisamide and the C_e/C_s ratio ($r = 0.7$, $P < 0.001$, data not shown) suggesting that serum albumin was responsible for zonisamide partitioning into the erythrocytes.

Several investigators have indicated that an increase in the unbound fraction is theoretically associated with corresponding increases in clearance and the volume of distribution of various drugs (Gibaldi & Koup 1981). In this study, significant increases in Vd and daily free fraction (AUC_f/AUC_s; Cloyd et al 1993) were observed on the day of the operation compared with the post-operative values. Zonisamide is bound primarily to erythrocytes rather than to plasma proteins; erythrocyte-borne drugs in-vivo are, however, available for distribution to tissues (Cornford & Landon 1985). A decrease in serum protein binding and an increase in uptake by erythrocytes, which could increase the volume of distribution, would have contributed to the fall in AUCs during the surgery. It remains unclear, however, why the state of surgery should influence the distribution of zonisamide. Factors such as initial water load and subsequent maintenance of the state of hydration might have led to an increase in the extracellular volume, and hence to an increase in the potential volume of drug distribution (Faruq et al 1990). In such cases, the observed serum drug concentration should also be lower, because the administered dose has to be distributed over a much larger volume.

Our studies on valproic acid and zonisamide disposition during and after brain surgery have illustrated that surgery is a factor influencing drug metabolism, renal excretion, and distribution. With valproic acid, we found that a reduction of intrinsic clearance was the predominant effect (Ieiri et al 1995), whereas with zonisamide in this study the renal excretion and accumulation in erythrocytes were significantly increased during surgery. Cornford & Landon (1985) studied the brain capillary transit of zonisamide and suggested that zonisamide in both plasma and erythrocytes could equilibrate across the blood-brain barrier in the course of a single transcapillary transit. Approximately one-half of the drug gaining access to the brain in a single transcapillary passage is erythrocyte-borne. Many workers have suggested that red blood cells, or even red cell ghosts, might serve as carriers of drugs and facilitate the delivery of agents to a desired site (Ihler 1983). It remains to be determined whether increased uptake of this AED by erythrocytes, thereby resulting in the drug gaining greater access to the desired brain site, would be of a prophylactic benefit to seizure patients.

Acknowledgements

We thank the resident physicians and the faculty of the Department of Neurosurgery, Kyushu University Hospital, for their enthusiastic cooperation. We thank S. Emoto and M. Inokuchi for their technical assistance throughout this study.

References

Boddy, A., Edwards, P., Rowland, M. (1989) Binding of sulfonamides to carbonic anhydrase: influence on distribution within blood and on pharmacokinetics. Pharm. Res. 6: 203–209

Casper, R., Garver, D. L., Dekirmenjian, H., Chang, S., Davis, J. M. (1980) Phenothiazine levels in plasma and red cells. Arch. Gen. Psychiatry 37: 301–306

Cloyd, J. C., Fischer, J. H., Kriel, R. L., Kraus, D. M. (1993) Valproic acid pharmacokinetics in children. Effect of age and antiepileptic drugs on protein binding and intrinsic clearance. Clin. Pharmacol. Ther. 53: 22–29

Cornford, E. M., Landon, K. P. (1985) Blood-brain barrier transport of CI-912: single-passage equilibration of erythrocyte-borne drugs. Ther. Drug Monit. 7: 247–254

Driessen, O., Treuren, L., Meijer, J. W. (1989) Distribution of drugs over whole blood: I. The transport function of whole blood for valproate. Ther. Drug. Monit. 11: 384–389

Evans, G. H., Shand, D. G. (1973) Disposition of propranolol. VI. Independent variation in steady-state circulating drug concentrations and half-life as a result of plasma drug binding in man. Clin. Pharmacol. Ther. 14: 494–500

Faruq, H., Noormohamed, W., Robin, M., Josh, J. D., Ariel, F. L. (1990) Renal responses and pharmacokinetics of piretanide in human: effect of route of administration, state of hydration and probenecid pre-treatment. J. Pharmacol. Exp. Ther. 254: 992–999

Funakoshi, S., Deutsch, H. F. (1968) Human carbonic anhydrases: I. Isolation and demonstration of isozymes in erythrocytes. J. Biol. Chem. 243: 6474–6481

Garver, D. L., Dekirmenjian, H., Davis, J. M., Casper, R., Ericksen, S. (1977) Neuroleptic drug levels and therapeutic response: preliminary observations with red blood cell bound butaperazine. Am. J. Psychiatry 134: 304–307

Gibaldi, M., Koup, J. R. (1981) Pharmacokinetic concepts-drug binding, apparent volume of distribution and clearance. Eur. J. Clin. Pharmacol. 20: 299–305

Hughes, I. E., Jellett, L. B., Ilett, K. F. (1976) The influence of various factors on the in vitro distribution of haloperidol in human blood. Br. J . Clin. Pharmacol. 3: 285–288

Ieiri, I., Morioka, T., Ichimiya, T., Nishio, S., Fukui, M., Ohtsubo, K., Higuchi, S. (1995) Pharmacokinetic study of valproic acid sustained-release preparation in patients undergoing brain surgery. Ther. Drug. Monit. 17: 6–11

Ihler, G. M. (1983) Erythrocyte carriers. Pharmacol. Ther. 20: 151–169

Ito, T., Yamaguchi, T., Miyazaki, H., Sekine, Y., Shimizu, M., Ishida, S., Yagi, K., Kakegawa, N., Seino, M., Wada, T. (1982) Pharmacokinetic studies of AD-810, a new antiepileptic compound. Arzneim. Forsch. 32: 1581–1586

Kurata, D., Wilkinson, G. R. (1974) Erythrocyte uptake and plasma binding of diphenylhydantoin. Clin. Pharmacol. Ther. 16: 355–362

Maren, T. H. (1976) Carbonic anhydrase: chemistry, physiology, and inhibition. Physiol. Rev. 47: 595–781

Matsumoto, K., Miyazaki, H., Fujii, T., Kagemoto, T., Maeda, T., Hashimoto, M. (1983) Absorption, distribution and excretion of 3-(sulfomoyl[^{14}C]methyl)-1,2-benzisoxazole (AD-810) in rats, dogs and monkeys and of AD-810 in men. Arzneim. Forsch. 33: 961–968

Matsumoto, K., Miyazaki, H., Fujii, T., Hashimoto, M. (1989a) Binding of sulfonamides to erythrocytes and their components. Chem. Pharm. Bull. 37: 1913–1915

Matsumoto, K., Miyazaki, H., Fujii, T., Amejima, H., Furukawa, H., Hashimoto, M. (1989b) Binding of sulfonamides to erythrocyte proteins and possible drug-drug interaction. Chem. Pharm. Bull. 37: 2807–2810

Nakasa, H., Komiya, M., Ohmori, S., Rikihisa, T., Kiuchi, M., Kitada, M. (1993) Characterization of human liver microsomal cytochrome P450 involved in the reductive metabolism of zonisamide. Mol. Pharmacol. 44: 216–221

Ojemann, L. M., Shastri, R. A., Wilensky, A. J., Friel, P. N., Levy, R. H. (1986) Comparative pharmacokinetics of CI-912 in epileptic patients on carbamazepine or phenytoin monotherapy. Ther. Drug Monit. 8: 293–296

Peters, D. H., Sorkin, E. M. (1993) Zonisamide. A review of its pharmacodynamic and pharmacokinetic properties, and therapeutic potential in epilepsy. Drugs 45: 760–787

Sackellares, J. C., Donofrio, P. D., Wagner, J. G., Abou-Khaalil, B., Berent, S., Aasued-Hoyt, K. (1985) Pilot study of zonisamide (1,2-benzisoxazole-3-methanesulphonamide) in patients with refractory partial seizures. Epilepsia 26: 206–211

Voltonen, M., Laitinen, J. T., Eriksson, L. (1993) Renal melatonin excretion in sheep is enhanced by water diuresis. J. Endocrinol. 138: 445–450

Wilensky, A. J., Friel, P. N., Ojemann, L. M., Almes, M. J., Levy, R. H., Buchanan, R. A. (1984) Pharmacokinetics of CI-912 in epileptic patients. In: Levy, R. H., Pitlick, W. H., Eichelbaum, M., Meijer, J. (eds) Metabolism of Antiepileptic Drugs, Raven Press, New York, pp 209–216

Wilensky, A. J., Friel, P. N., Ojemann, L. M., Dodrill, C. B., McKormaick, K. B., Levy, R. H. (1985) Zonisamide in epilepsy: a pilot study. Epilepsia 26: 212–220