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The pharmacokinetic inter-relationship of tiagabine in blood, cerebrospinal fluid and brain extracellular fluid (frontal cortex and hippocampus)

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KEYWORDS

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Summary Purpose: Tiagabine is a unique antiepileptic drug with a novel mechanism of action. Whilst some limited data are available as to the peripheral blood pharmacokinetics of tiagabine, data regarding the kinetics of tiagabine in the central brain compartment are very limited. We therefore sought to investigate serum, cerebrospinal fluid (CSF) and frontal cortex and hippocampal extracellular fluid (ECF) kinetic inter-relationship of tiagabine in a freely moving rat model. **Methods:** Adult male rats were implanted with either a jugular vein catheter and a cisterna magna catheter for blood and CSF sampling, respectively, or a blood catheter and a microdialysis probe in the hippocampus and frontal cortex (for ECF sampling). Tiagabine was administered intraperitoneal (i.p.) at 20 or 40 mg/kg and blood, CSF and ECF were collected at timed intervals for the measurement of tiagabine concentrations by high performance liquid chromatography. **Results:** Tiagabine concentrations in blood and CSF rose linearly and dose-dependently and time to maximum concentration (T_{max}) was 15 and 29 min, respectively. Mean CSF/serum tiagabine concentration ratios (range, 0.008–0.01) were much smaller than the mean free/total tiagabine concentration ratios in serum (0.045 ± 0.003). Entry of tiagabine into brain ECF (frontal cortex and hippocampus) was rapid with T_{max} values of 31–46 min. Distribution of tiagabine in brain was not brain region specific with values in the frontal cortex and hippocampus being indistinguishable. Whilst elimination from CSF was comparable to that of serum, half-life ($t_{1/2}$) values in ECF were three times longer. **Conclusions:** Tiagabine is associated with linear kinetic characteristics and with rapid brain penetration. However, CSF concentrations are not reflective of free non-protein-bound concentrations in serum. The observation that tiagabine elimination from the brain is threefold slower than that seen in blood, may explain as to the relatively long duration of action of tiagabine.

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Introduction

Tiagabine ([*R*-(–)-*N*-(4,4-di-(3-methylthien-2-yl)-but-3-enyl) nipecotic acid] hydrochloride) is a unique antiepileptic drug (AED) with a novel mechanism of action in that it selectively and specifically inhibits the uptake of γ -aminobutyric acid (GABA) into astrocytes and neurones, by the transporter GAT-1, and thus increases the extracellular concentration of GABA in the brain. Tiagabine is effective in the management of partial seizures with or without secondary generalisation, and is licensed for use as adjunctive therapy in patients refractory to available first-line AEDs.^{1–4}

Tiagabine is effective in a variety of seizure models including amygdala-kindled seizures,⁵ audiogenic seizures⁶ and convulsions associated with status epilepticus induced in cobalt-lesioned rats by the administration of homocysteine thiolactone.⁷ The latter observation raises the possibility that it may be useful in the management of status epilepticus in patients. However, from a pharmacokinetic aspect, drugs used in the management of status epilepticus need to be able to rapidly and efficiently penetrate the blood–brain barrier and to have an elimination half-life that does not require frequent dosing.

Whilst data on the blood pharmacokinetics of tiagabine in man are abundant,⁸ such data are scarce in the rat.⁹ Furthermore, data regarding the kinetics of tiagabine in the brain are very limited indeed; one study has reported on whole brain tiagabine concentrations at 1 h after intraperitoneal administration of tiagabine.¹⁰ Tiagabine concentrations increased dose-dependently and were higher in the dorsal cortex compared to those measured in the cerebellum. Some additional information on the central brain kinetics of tiagabine can be deduced indirectly via observation of central GABA changes. Thus, tiagabine dose-dependently increased GABA extracellular fluid (ECF) concentrations two- to threefold in the globus pallidus, ventral pallidum and in the substantia nigra of rats.¹¹ However, tiagabine had no effect on cerebrospinal fluid (CSF) GABA concentrations after 2 days of tiagabine administration (50–200 mg/kg/day).¹² A microdialysis study of a single patient with epilepsy, reported an ~50% increase in GABA ECF concentrations at 1 h after a single dose of oral tiagabine ingestion and the increase was sustained for several hours.¹³

The present study sought to ascertain the peripheral blood pharmacokinetic and central brain (CSF and ECF) kinetic inter-relationship of tiagabine using a well established and validated rat model.^{14–16}

Methods

Animals

Male Sprague–Dawley rats weighing 250–300 g (A. Tuck & Son Ltd., Battlesbridge, Essex, UK) were group-housed in contiguous cages and fed ad libitum on normal laboratory diet (SDS R and M number 1 expanded, Scientific Dietary Services, Witham, Essex, UK) and water. A 12-h light–dark cycle (lights on 06:00 h) and ambient temperature of 25 °C were maintained. All the rats were allowed to acclimatise to their new environment for at least 3 days prior to experimentation. All animal procedures strictly followed Home Office regulations and were performed under the Animal (Scientific Procedures) Act 1986.

Surgical procedures, blood, CSF and microdialysis sampling

Rats were anaesthetised with 2% halothane (Merial Animal Health Ltd., Dublin, Ireland) inhalation. Two surgical procedures were undertaken. In the first group of rats, catheters were implanted in the right jugular vein for blood sampling and in the cisterna magna for CSF sampling as previously described.¹⁴ For the second group of rats the procedure involved the implantation of a catheter in the right jugular vein for blood sampling and a microdialysis probe in both the hippocampus (from bregma 5.6 mm posterior, 5 mm lateral, 8.2 mm ventral) and the frontal cortex (from bregma 2.5 mm anterior, 1.5 mm lateral, 5.5 mm ventral) for monitoring of the ECF. Stereotaxic placement of the probes was according to the atlas of Paxinos and Watson.¹⁷ Post-surgery rats were housed singly in perspex cages.

Two days later, when animals had fully recovered, the microdialysis probes and the CSF and blood catheters were checked for patency. Animals with blood and CSF catheters were investigated as follows: 20 or 40 mg/kg tiagabine was administered by intraperitoneal (i.p.) injection. Blood (200 μ l) and CSF (20 μ l) were sampled via the implanted catheters at timed intervals over a 7-h period (15, 30, 45, 60, 90, 120, 150, 180, 240, 300, 360, 390 and 420 min). In order to prevent the development of hypovolemia, an equivalent volume of heparinized saline was administered after each blood sampling. Blood samples were collected in 0.5 ml polyethylene tubes (Treff AG, Switzerland), vortex mixed and centrifuged at 10,000 rpm for 5 min (Abbott micro-centrifuge, Abbott Diagnostics, Maidenhead, UK). Supernatant serum and CSF were

stored frozen (-70°C) until analysis for tiagabine content.

Animals with blood catheters and microdialysis probes were investigated as follows: Artificial CSF (CSF composition (in mM): NaCl 125, KCl 2.5, MgCl_2 1.18 and CaCl_2 1.26) was perfused through the probes at $2\ \mu\text{l}/\text{min}$. A baseline sample of blood ($200\ \mu\text{l}$) and three baseline dialysate samples ($20\ \mu\text{l}$) were taken before drug administration. The rats were then injected i.p. with $40\ \text{mg}/\text{kg}$ tiagabine and venous blood samples ($200\ \mu\text{l}$) were withdrawn at 10, 20, 30, 40, 50, 60, 75, 90, 105 and 120 min. Dialysate samples ($20\ \mu\text{l}$) were collected at 10-min intervals for the first 60 min and at 15-min intervals for the subsequent 60-min period and sampling time was corrected for dead-space volume time. Blood and microdialysate samples were collected in $0.5\ \text{ml}$ polyethylene tubes (Treff AG). Blood samples were prepared as described above and supernatant serum and microdialysate were stored frozen (-70°C) until analysis for tiagabine content.

Microdialysis probe construction and in vitro recovery

Concentric microdialysis probes with filtral 12 (Hospal, Rugby, UK) dialysis membrane ($4\ \text{mm}$ long, $200\ \mu\text{m}$ diameter) and internal vitreous silica tubing (SGE, Milton Keynes, UK) were prepared as previously described. In vitro probe recovery was determined by placing each microdialysis probe into a beaker containing a solution of $200\ \text{nmol}/\text{l}$ tiagabine, constituted in artificial CSF, and then perfusing with artificial CSF at 37°C with a flow rate of $2\ \mu\text{l}/\text{min}$. Samples ($40\ \mu\text{l}$) were collected every 20 min for 120 min and stored at -70°C until analysis for tiagabine content.

Measurement of tiagabine concentration

Tiagabine concentrations in serum, CSF and microdialysates were measured by high performance liquid chromatography (HPLC). The HPLC system comprised: a Gilson 307 pump, a Gilson 234 autoinjector, ESA coulochem electrochemical detector and an analytical cell (model 5011). A Hyper-sil BDS-C18, $3\ \mu\text{m}$, $125\ \text{mm} \times 3\ \text{mm}$ column (Hewlett Packard, Stockport, Cheshire, UK) and LiChrosphere select B 4×4 ($5\ \mu\text{m}$) pre-column (Hewlett Packard) were used. Chromatograms were collated using Unipoint System Software (Version 1.71). The lower limit of quantification for tiagabine was $0.5\ \text{nmol}/\text{l}$ (CV, 6%). The lower limit of detection was $0.1\ \text{nmol}/\text{l}$. The procedure used for the determination of the free non-protein-bound serum

tiagabine concentration was the same as for total tiagabine concentrations except that samples were first filtered through an Amicon Centrifree Micropartition System (Amicon, Stonehouse, UK) using a Sigma 2K15 centrifuge with a temperature setting of 25°C .

Kinetic and statistical analysis

Pharmacokinetic parameters were computed (Microsoft Excel) based on a one-compartment model for CSF and microdialysate and a two-compartment model for blood with first order elimination. The following pharmacokinetic parameters were determined: time to maximal concentration (T_{max}), maximal concentration (C_{max}), terminal elimination half-life ($t_{1/2}$) and area under the concentration versus time curve (AUC). The AUC was calculated by linear trapezoid summation and extrapolation to infinite time as appropriate. The K_e was estimated by least-square regression analysis of the terminal log-linear portion of the plasma concentration versus time profile. The ($t_{1/2}$) was calculated by dividing the natural log of 2 by K_e . C_{max} and T_{max} were read directly from the concentration versus time curves.

Non-parametric Mann–Whitney tests were used to ascertain significance and a P value of 0.05 was considered statistically significant.

Results

Blood pharmacokinetics

Tiagabine pharmacokinetic parameters in serum after 20 and $40\ \text{mg}/\text{kg}$ tiagabine administration are shown in Table 1. Tiagabine concentrations rose dose-dependently and C_{max} values were achieved at the time of first sampling (15 min post-dose). Concentrations subsequently declined rapidly and the decline was exponential (Fig. 1). Both C_{max} and AUC values increased linearly and dose-dependently. Tiagabine $t_{1/2}$ values were $55 \pm 2.3\ \text{min}$ and $50 \pm 2.6\ \text{min}$ after 20 and $40\ \text{mg}/\text{kg}$ administration, respectively, and were not statistically different ($P > 0.05$, Mann–Whitney test). At 240 min and at subsequent time points tiagabine concentrations were just quantifiable and were of the order of $150\ \text{nmol}/\text{l}$.

The free non-protein-bound tiagabine concentration was determined in serum. At 15 min post-dose (the time of first sample), the tiagabine free fraction was 1.4%. However, subsequent samples ($n = 8$) showed a larger free fraction value ($4.5 \pm 0.3\%$) and were both time and concentration independent.

Table 1 Pharmacokinetic parameters of tiagabine in serum, CSF and brain ECF after tiagabine administration at 20 and 40 mg/kg ($n = 5-7$).

	T_{max} (min)	C_{max} (nmol/l)	AUC (nmol h/l)	$t_{1/2}$ (min)
Serum				
Tiagabine (20 mg/kg)	15 ± 0	14905 ± 1733	9439 ± 909	55 ± 2.3
Tiagabine (40 mg/kg)	16 ± 0.3	27754 ± 2000	25147 ± 3491	50 ± 2.6
CSF				
Tiagabine (20 mg/kg)	28 ± 0.7	49 ± 6.7	51 ± 7.6	40 ± 2.6
Tiagabine (40 mg/kg)	32 ± 0.9	95 ± 5.4	138 ± 13	64 ± 2.7
ECF				
Tiagabine (40 mg/kg)				
Frontal cortex	41 ± 5	35 ± 1	154 ± 25	174 ± 32
Hippocampus	34 ± 3	38 ± 2	134 ± 10	133 ± 9

All values are mean ± S.E.M. T_{max} : time to maximal concentration; C_{max} : maximal concentration; AUC: area under the concentration vs. time curve; $t_{1/2}$: terminal elimination half-life; CSF: cerebrospinal fluid; ECF: extracellular fluid.

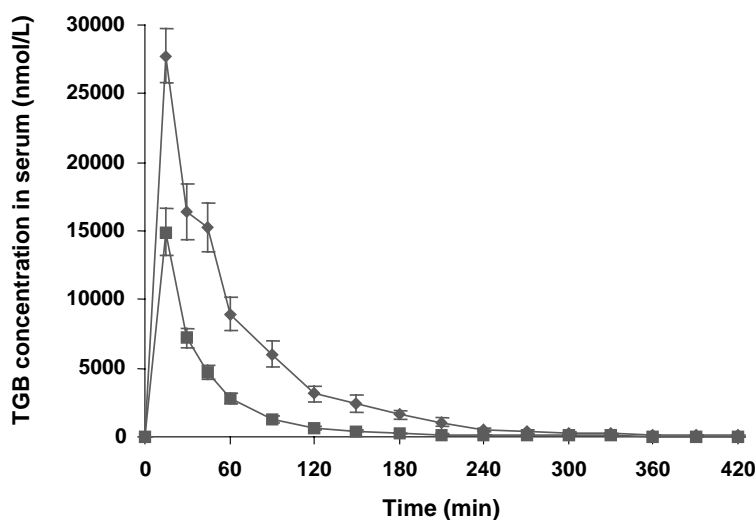


Figure 1 Tiagabine (TGB) concentration vs. time profiles in serum after 20 mg/kg (■) and 40 mg/kg (◆) TGB administration. Values are mean ± S.E.M. ($n = 5-7$).

Neuropharmacokinetics

CSF

The kinetic constants after 20 and 40 mg/kg tiagabine administration in CSF are shown in Table 1. After drug administration, tiagabine rapidly and readily penetrated into the CSF compartment and tiagabine was detectable in CSF at the time of first sampling (15 min; Fig. 2). Mean T_{max} values varied between 28 and 32 min and were dose-independent. However, compared to serum, CSF T_{max} values were double the value (30 min versus 15 min). C_{max} and AUC values increased linearly and dose-dependently. In contrast to serum CSF $t_{1/2}$ values were significantly longer after 40 mg/kg

tiagabine administration (64 ± 2.7 min) compared to when 20 mg/kg tiagabine was administered (40 ± 2.6 min; $P < 0.01$, Mann-Whitney test).

Fig. 3 shows the tiagabine concentration ratio of CSF/serum over time after 40 mg/kg tiagabine administration. There was a tendency towards equilibration (as determined by a constant CSF/serum tiagabine concentration ratio) by 30 min post-tiagabine administration. Rapid equilibration was also observed after administration of 20 mg/kg tiagabine (data not shown). Mean CSF/serum ratios were 0.008 ± 0.0006 after 20 mg/kg tiagabine administration and 0.01 ± 0.0009 after 40 mg/kg tiagabine administration. These values are not statistically different ($P > 0.05$, Mann-Whitney test).

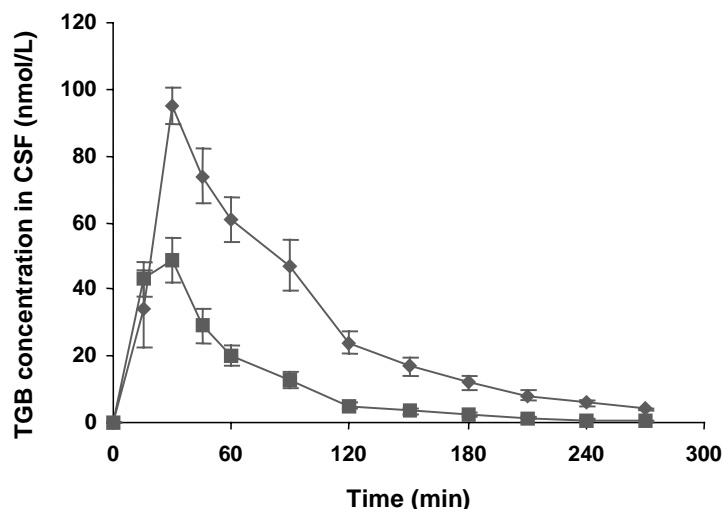


Figure 2 Tiagabine (TGB) concentration vs. time profiles in CSF after 20 mg/kg (■) and 40 mg/kg (◆) TGB administration. Values are mean \pm S.E.M. ($n = 5-7$).

Extracellular fluid kinetics

The mean \pm S.E.M in vitro recovery for tiagabine for all the microdialysis probes was $10 \pm 0.3\%$ at a dialysate flow rate of $2 \mu\text{l}/\text{min}$. The artificial CSF solution used contained $200 \text{ nmol}/\text{l}$ tiagabine and these recovery data were used to adjust the in vivo ECF concentration data.

The pharmacokinetic parameters of tiagabine in brain ECF frontal cortex and hippocampus after 40 mg/kg tiagabine administration are shown in Table 1. The corresponding tiagabine concentration versus time profiles are shown in Fig. 4. After drug administration, blood-brain barrier penetration was rapid and tiagabine was detectable at the time of first sampling (10 min post-dose). Mean T_{max} values for frontal cortex and hippocampus were $41 \pm 5 \text{ min}$ and $34 \pm 3 \text{ min}$, respectively. AUC values

were comparable for frontal cortex and hippocampus ($154 \pm 25 \text{ nmol h}/\text{l}$ versus $134 \pm 10 \text{ nmol h}/\text{l}$, respectively). Overall, there were no statistically significant difference in tiagabine kinetic parameters between brain frontal cortex and hippocampus ($P > 0.05$, Mann-Whitney test). However, the half-life values in ECF frontal cortex and hippocampus were significantly longer (up to three times) than that seen in the CSF and in blood.

Fig. 5 shows the tiagabine concentration ratio of ECF/serum for frontal cortex and hippocampus over time after 40 mg/kg tiagabine administration. In contrast to CSF, there was no tendency towards equilibration (as determined by a rising ECF/serum tiagabine concentration ratio) during the study period of up to 120 min. However, the mean ECF/serum ratios for frontal cortex

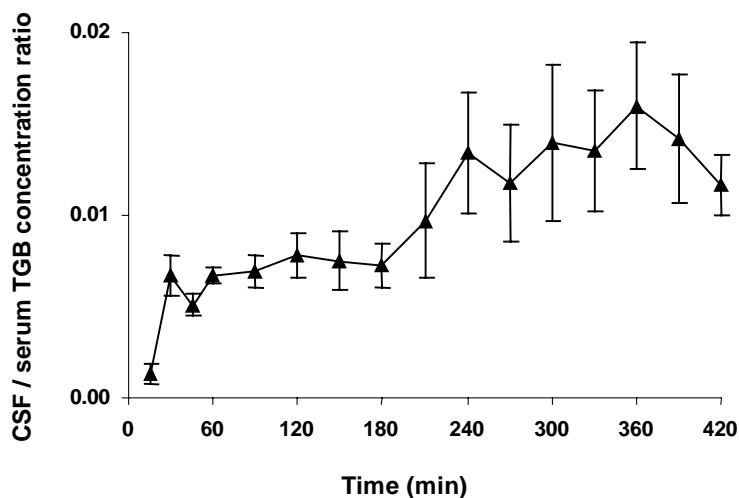


Figure 3 CSF/serum tiagabine (TGB) concentration ratios vs. time after 40 mg/kg TGB administration. Values are mean \pm S.E.M. ($n = 5$).

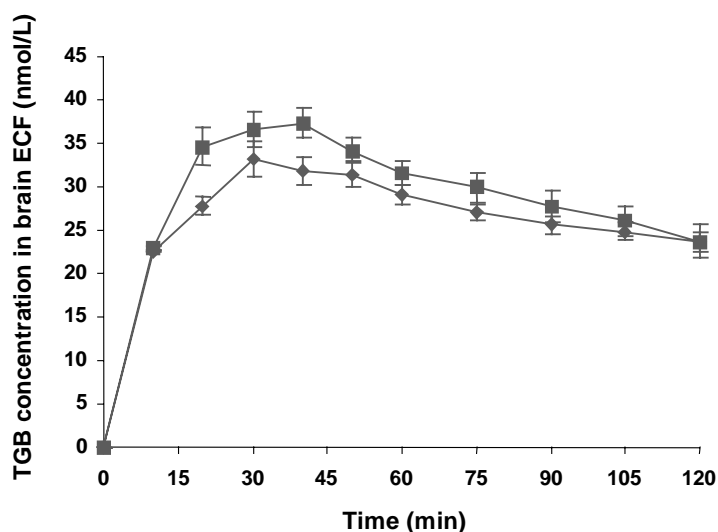


Figure 4 Tiagabine (TGB) concentration vs. time profiles in frontal cortex (◆) and hippocampal (■) ECF after 40 mg/kg TGB administration. Values are mean \pm S.E.M. ($n = 5-7$).

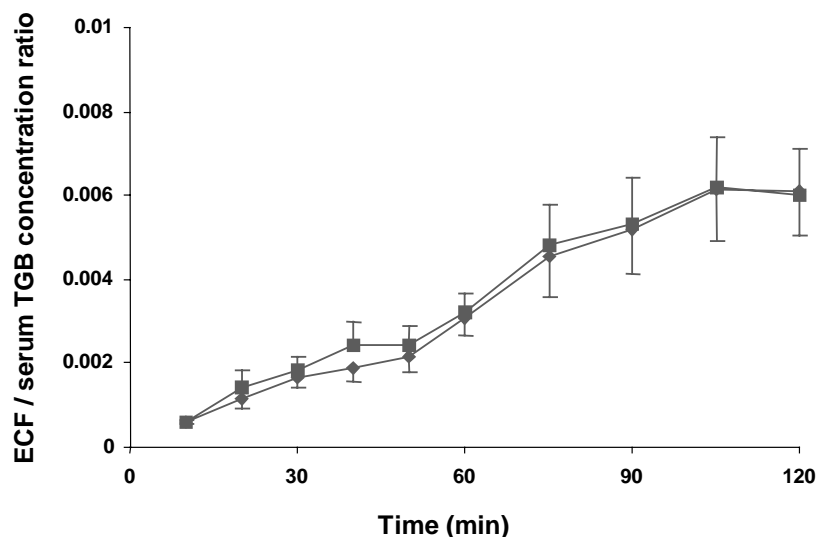


Figure 5 ECF/serum tiagabine (TGB) concentration ratios vs. time profiles in frontal cortex (◆) and hippocampal (■) after 40 mg/kg TGB administration. Values are mean \pm S.E.M. ($n = 5-7$).

and hippocampus were indistinguishable ($P > 0.05$, Mann-Whitney test).

Discussion

This is the first study to investigate the temporal pharmacokinetic inter-relationship of tiagabine in blood, CSF and brain ECF. The major findings of this study are: (1) the pharmacokinetics of tiagabine in serum are linear and dose-dependent; (2) tiagabine is substantially protein bound (95%) in serum; (3) the elimination of tiagabine from serum is rapid ($t_{1/2}$ values, 50–55 min) and independent of dose; (4) tiagabine rapidly and readily penetrated the

blood–brain barrier and was detectable within minutes in both CSF and brain ECF; (5) although the CSF pharmacokinetics of tiagabine paralleled that seen in serum, CSF concentrations did not reflect free drug concentrations in serum; (6) whilst tiagabine $t_{1/2}$ values were comparable in CSF and serum, values were three times larger in frontal cortex and hippocampal ECF; (7) tiagabine distribution in brain frontal cortex and hippocampal ECF were indistinguishable.

Although there is a lack of published studies describing the kinetics of tiagabine in the rat, the sparse data reported by Suzdak and Jansen⁹ are generally in agreement with those described in the present report. After oral ingestion of an

unspecified tiagabine dose, tiagabine T_{\max} values were ~ 30 min;⁹ slightly longer, as might be expected, compared to after i.p. administration in the present study (Table 1). Tiagabine serum protein binding was also comparable (92–93% versus 95–96%, this study). That in the present study ECF tiagabine $t_{1/2}$ values were three times longer than that observed in the blood compartment (Table 1), corroborates the indirect data presented by Suzdak and Jansen.⁹ It was noted that although after [¹⁴C]tiagabine administration (dose and route of administration were not reported) the decrease in tissue concentrations of [¹⁴C]tiagabine (liver, kidney and lung) followed the decrease in serum concentrations, the brain/plasma ratio remained constant. This was interpreted by the authors as indicating a slower elimination of tiagabine from the brain or that equilibration between serum and brain is slow. Indeed, the data presented in the present study would suggest that both these processes might pertain (Table 1 and Fig. 5). Furthermore, the CSF $t_{1/2}$ values reported in the present study were significantly larger at the higher tiagabine dose (40 mg/kg; 64 ± 2.7 min) when compared to values obtained at the lower dose (20 mg/kg; 40 ± 2.6 min). These data corroborate the observation that elimination of tiagabine from the brain was dose-dependent.⁹ Lastly, the observation that the distribution of tiagabine in the frontal cortex and hippocampus is similar is in agreement with the *in vitro* tissue binding studies with [³H]tiagabine.¹⁸

The rate of penetration of drugs, including AEDs, into CSF and brain is considered to be determined essentially by three physicochemical characteristics, namely degree of ionisation, lipid solubility and degree of serum protein binding. In relation to AEDs, lipid solubility appears to be the major determinant of rate of entry into the CSF compartment.¹⁹ Because tiagabine has high lipid solubility it would be expected to gain rapid entry into the brain and indeed this was the case as tiagabine was detectable in both CSF and frontal cortex and hippocampal ECF at the time of first sampling (10–15 min post-dose). Furthermore, the rate of entry into the CSF was not dose-dependent, as there was no difference in T_{\max} values after different tiagabine dose administration (20 and 40 mg/kg). Thus, tiagabine transportation across the blood–CSF barrier in the choroid plexus was not rate-limiting at the concentration ranges achieved. This characteristic may very well apply to transportation of tiagabine across the blood–brain barrier but since the kinetics of tiagabine in frontal cortex and hippocampal ECF were determined after only one tiagabine dose (40 mg/kg) this cannot be definitively conclude from the present data.

Free (non-protein-bound) drug concentrations in serum are often used as a reflection of CNS drug concentrations. In the present study, mean CSF/serum tiagabine concentration ratios (range, 0.008–0.01) are much smaller than the mean free/total tiagabine concentration ratios in serum (0.045 ± 0.003). These data suggest that free tiagabine serum concentrations do not accurately reflect CSF tiagabine concentrations. This is in contrast to other AEDs (e.g. carbamazepine, phenytoin, primidone, phenobarbitone and lamotrigine) studied in this animal model where CSF concentrations do reflect free drug concentrations in serum and emphasises the importance of studying the central neuropharmacokinetics of a drug at the site of drug action.^{14,16,20,21}

That tiagabine elimination from the brain is much slower than that that occurs in serum could be of significant clinical importance since it would suggest that the pharmacological effect of tiagabine is much longer than that that would be suggested by its blood pharmacokinetic profile. Thus, the short tiagabine $t_{1/2}$ in serum, which is generally perceived as a disadvantage for the clinical therapeutics of tiagabine,¹⁶ may actually not be as relevant if indeed the data observed in the present study could be extrapolated to man. A further consideration in relation to the difference between the central and peripheral kinetics of tiagabine would be that if the dosing strategy for tiagabine is based on its serum kinetics, tiagabine would accumulate in the brain. Thus, if non-convulsive status epilepticus, that has been reported in patients treated with tiagabine,^{22–24} is the consequence of GABAergic hyperfunction, as has been suggested,²⁵ then it could be the consequence of tiagabine accumulation in the brain.

The tiagabine doses used in the present study (20 and 40 mg/kg) are higher than the ED_{50} for tiagabine in animal seizure models²⁶ or indeed doses that are used clinically.⁴ Because tiagabine is highly protein bound (95–96%), only a very small proportion (4–5%) of the tiagabine concentration achieved in blood passes the blood–brain barrier to enter the CSF and ECF compartments. Consequently, dose selection was based on the rationale and the need to achieve quantifiable tiagabine concentrations in CSF and ECF. That the doses used in the present study do not impede on the value of the data collected, is evidenced by the fact that the overall conclusions reached are similar to those described by Suzdak and Jansen.⁹

Finally, the regional distribution of tiagabine in the brain needs some comment. The present study clearly shows that the distribution of tiagabine in frontal cortex and hippocampal ECF is almost

identical and the data corroborate data from in vitro tissue binding studies with [³H]tiagabine.¹⁸ GABA transporters (designated GAT-1, GAT-2, GAT-3 and BGT-1 according to their pharmacological sensitivity)²⁷ have a differential anatomical distribution throughout the nervous system.^{28,29} As GAT-1 is primarily located in the cerebral cortex and hippocampus and as tiagabine (through its specificity for GAT-1) is equally distributed in these brain regions, it would be expected the pharmacological action of tiagabine would be brain region specific.

In conclusion, this study has shown that tiagabine has linear peripheral and central kinetics and with rapid brain and CSF penetration although CSF concentrations do not reflect free drug concentrations in serum. Its distribution in brain cerebral cortex and hippocampus would suggest it is not region specific and its elimination from the brain greatly outlasts that seen in blood.

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References

- Sachdeo RC, Leroy RF, Krauss GL, et al. Tiagabine-therapy for complex partial seizures: a dose-frequency study. *Arch Neurol* 1997;54:595–601.
- Kalviainen R, Brodie MJ, Duncan J, Chadwick D, Edwards D, Lyby K. A double blind placebo-controlled trial of tiagabine given three times daily as add-on therapy for refractory seizures. *Epilepsy Res* 1998;30:31–40.
- Uthman BM, Rowan AJ, Ahmann PA, et al. Tiagabine for complex partial seizures: a randomized, add-on, dose-response trial. *Arch Neurol* 1998;55:56–62.
- Gaitatzis A, Patsalos PN, Sander JWAS. The clinical efficacy of tiagabine. *Rev Contemp Pharmacother* 2002;12:235–50.
- Dalby NO, Nielsen EB. Tiagabine exerts an anti-epileptogenic effect in amygdala kindling epileptogenesis in the rat. *Neurosci Lett* 1997;229:135–7.
- Faingold CL, Randall ME, Anderson CA. Blockade of GABA uptake with tiagabine inhibits audiogenic seizures and reduces neuronal firing in the inferior colliculus of the genetically epilepsy-prone rat. *Exp Neurol* 1994;126:225–32.
- Walton NY, Gunawan S, Treiman DM. Treatment of experimental status epilepticus with the GABA uptake inhibitor tiagabine. *Epilepsy Res* 1994;19:237–44.
- Wang X, Patsalos PN. The pharmacokinetic profile of tiagabine. *Rev Contemp Pharmacother* 2002;12:225–33.
- Suzdak PD, Jansen JA. A review of the preclinical pharmacology of tiagabine: a potent and selective anticonvulsant GABA uptake inhibitor. *Epilepsia* 1995;36:612–26.
- Sills GJ, Patsalos PN, Butler E, Forrest G, Ratnaraj N, Brodie MJ. Visual field constriction: accumulation of vigabatrin but not tiagabine in the retina. *Neurology* 2001;57:196–200.
- Fink-Jensen A, Suzdak PD, Swedberg MD, Judge ME, Hansen L, Nielsen PG. The gamma-aminobutyric acid (GABA) uptake inhibitor, tiagabine, increases extracellular brain levels of GABA in awake rats. *Eur J Pharmacol* 1992;220:197–201.
- Halonen T, Nissinen J, Jansen JA, Pitkanen A. Tiagabine prevents seizures neuronal damage and memory impairment in experimental status epilepticus. *Eur J Pharmacol* 1996;299:69–81.
- During M, Mattson R, Scheyer R. The effect of tiagabine HCl on extracellular GABA levels in the human hippocampus. *Epilepsia* 1992;33(Suppl 3):83.
- Patsalos PN, Alavijeh MS, Semba J, Lolin YI. A freely moving and behaving rat model for the chronic and simultaneous study of drug pharmacokinetics (blood) and neuropharmacokinetics (cerebrospinal fluid): haematological and biochemical characterisation and kinetic evaluation using carbamazepine. *J Pharmacol Toxicol Methods* 1992;28:21–8.
- Tong X, Patsalos PN. A microdialysis study of the novel antiepileptic drug levetiracetam: extracellular pharmacokinetics and effect on taurine in rat brain. *Br J Pharmacol* 2001;133:867–74.
- Wang X, Patsalos PN. A comparison of central brain (cerebrospinal and extracellular fluids) and peripheral blood kinetics of phenytoin after intravenous phenytoin or fosphenytoin. *Seizure* 2003;12:330–6.
- Paxinos G, Watson C. *The rat brain in stereotaxic coordinates*. 2nd ed. San Diego: Academic Press; 1986.
- Suzdak PD, Foged C, Anderson KE. Quantitative autoradiographic characterisation of the binding of [³H]tiagabine (NNC 05-328) to the GABA uptake carrier. *Brain Res* 1994;647:231–41.
- Loscher W, Frey HH. Kinetics of penetration of common antiepileptic drugs into cerebrospinal fluid. *Epilepsia* 1984;25:346–52.
- Nagaki S, Ratnaraj N, Patsalos PN. Blood and cerebrospinal fluid pharmacokinetics of primidone and its primary pharmacologically active metabolites, phenobarbital and phenylethylmalonamide in the rat. *Eur J Drug Metab Pharmacokinetics* 1999;24:255–64.
- Walker MC, Tong X, Perry H, Alavijeh MS, Patsalos PN. Comparison of serum, cerebrospinal fluid and brain extracellular fluid pharmacokinetics of lamotrigine. *Br J Pharmacol* 2000;130:242–8.
- Schapel G, Chadwick D. Tiagabine and non-convulsive status epilepticus. *Seizure* 1996;5:153–6.
- Eckardt KM, Steinhoff BJ. Nonconvulsive status epilepticus in two patients receiving tiagabine treatment. *Epilepsia* 1998;39:671–4.
- Ettinger AB, Bernal OG, Andriola MR, et al. Two cases of non-convulsive status epilepticus in association with tiagabine therapy. *Epilepsia* 1999;40:1159–62.
- Solomon GE, Labar D. Hypothesis that tiagabine-induced NCSE is associated with GABAergic hyperfunction, with GABA(B) receptors playing a critical role, is supported by a case of generalised NCSE induced by baclofen. *Epilepsia* 1998;39:1383.
- Walker MC. The mechanism of action of tiagabine. *Rev Contemp Pharmacother* 2002;12:213–23.
- Borden LA, Smith KE, Hartig PR, Branchek TA, Weinschank RL. Molecular heterogeneity of the γ -aminobutyric acid (GABA) transport system. *J Biol Chem* 1992;267:21098–104.
- Durkin MM, Smith KE, Borden LA, Weinschank RL, Branchek TA, Gustafson EL. Localisation of messenger RNAs encoding three GABA transporters in rat brain: an in situ hybridization study. *Mol Brain Res* 1995;33:7–21.
- Ribak CE, Tong WMY, Brecha NC. The GABA plasma membrane transporters, GAT-1 and GAT-3, display different distribution in the rat. *J Comp Neurol* 1995;367:595–606.