Comparisons of pharmacokinetic parameters of valproic acid between responsive and resistant adult epileptic

Duangchit Panomvana* Lakkana Boonmark*

Tatta Sriboonruang*   Somchai Towanabut**

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Objectives   :   *To determine whether there are valproic acid (VPA) pharmacokinetic (PK) parameters differences between the VPA-responsive and VPA-resistant patient groups.*

Methods   :   *Thirty-three subjects (14 patients were in VPA-resistant group and 19 patients in VPA-responsive group) who were epileptic patients were recruited from the Neurology Clinic, Neurology Institute, Bangkok. Their ages were 15 - 65 years old. They were either treated with VPA (Depakine Chrono) monotherapy or polytherapy in which the combined drug did not have the same metabolic pathways as VPA. At steady state condition, two blood samples were collected from the subjects during elimination phase, approximately five hours apart. Individual PK parameter values of VPA were estimated using the equations for a linear one-compartment model with a first-order declining of drug concentration after the end of 14 hours zero-order drug release. Chi-square test and the odds ratio were used to analize the data.*

* Department of Pharmacy Practice, Faculty of Pharmaceutical Sciences, Chulalongkorn University.

** Department of Medicine, Neurological Institute, Bangkok, Thailand.

Results   : *Under steady state condition after consuming Depakine Chrono®, a volume of distribution value (Vd) obtained from total VPA concentrations was statistically significantly different between two patient groups (p<0.05). Additionally, Vd value of total VPA less than 0.3 l/kg was associated with nonresponsive to VPA treatment (p<0.05). While, half life (t½) of total VPA less than 25 hours and parameters obtained from unbound VPA concentrations such as Vd value less than 0.7 l/kg and t½ less than 10 hours had higher tendency to belong to the VPA-resistant group but they did not reach statistical significance (p>0.05); however, all these parameters were taken significant when p <0.1.*

Conclusions   : *The PK parameter values estimated from two blood samples collected during the routine clinical therapeutic drug monitoring might be used to identify whether or not the epileptic patient responds to the VPA therapy and thus could reduce the steps of trial and error.*

Keywords   : *Valproic acid, Pharmacokinetic parameters, VPA-responsive patient, VPA-resistant patient.*

วัตถุประสงค์ : *เพื่อศึกษาเปรียบเทียบพารามิเตอร์ทางเภสัชจลนศาสตร์ของยาวาลโปรอิค แอซิด ระหว่างกลุ่มผู้ป่วยโรคลมชักที่ไม่ตอบสนองต่อการรักษา และผู้ป่วยที่ตอบสนองการรักษาด้วยวาลโปรอิค แอซิด*

วิธีการศึกษา : *ผู้ป่วยโรคลมชัก จำนวน 33 ราย (แบ่งเป็นผู้ป่วยไม่ตอบสนองต่อยาจำนวน 14 ราย และผู้ป่วยที่ตอบสนองต่อยา 19 ราย) ณ สถาบันประสาทวิทยา กทม. อายุระหว่าง 15 - 65 ปี ที่ได้รับการรักษาด้วยวาลโปรอิค แอซิดแบบเดี่ยวหรือแบบร่วมกับยาอื่นที่ไม่ส่งผลต่อเภสัชจลนศาสตร์ของวาลโปรอิค แอซิด ยินยอมเข้าร่วมการศึกษา โดยผู้ป่วยจะถูกเก็บตัวอย่างยาในเลือด 2 จุดหลังรับประทานยาจนถึงภาวะคงที่ของระดับยาในเลือดแล้ว โดยเจาะเลือดสองจุดห่างกัน ประมาณ 5 ชั่วโมง ณ ช่วงเวลาที่มีการกำจัดยา และนำค่าระดับยาในเลือดมาคำนวณพารามิเตอร์ทางเภสัชจลนศาสตร์ของยาวาลโปรอิค แอซิด และนำผลที่ได้มาวิเคราะห์ทางสถิติ โดยใช้ ไคว์สแคว์*

ผลการศึกษา : *พบปริมาตรการกระจายยาของวาลโปรอิค แอซิด (total VPA) มีความแตกต่างกันระหว่างผู้ป่วยสองกลุ่มอย่างมีนัยสำคัญทางสถิติ โดยพบความสัมพันธ์ของการไม่ตอบสนองต่อยาและปริมาตรการกระจายยาของวาลโปรอิค แอซิด ที่มีค่าน้อยกว่า 0.3 ลิตรต่อกิโลกรัม อย่างมีนัยสำคัญทางสถิติ นอกจากนั้นในกลุ่มผู้ป่วยที่ไม่ตอบสนองต่อยา พบแนวโน้มของความสัมพันธ์กับ ค่าครึ่งชีวิตของยาวาลโปรอิคแอซิด (total VPA) ที่มีค่าน้อยกว่า 25 ชั่วโมง ปริมาตรการกระจายตัวรูปแบบยาอิสระ (Unbound VPA) น้อยกว่า 0.7 ลิตรต่อกิโลกรัม และค่าครึ่งชีวิตของยาอิสระน้อยกว่า 10 ชั่วโมงแต่ไม่พบนัยสำคัญทางสถิติ*

สรุปผลการศึกษา : *ค่าพารามิเตอร์ทางเภสัชจลนศาสตร์จากผลการเจาะติดตามระดับยาในเลือดอาจสามารถนำมาทำนายผลการตอบสนองต่อยาวาลโปรอิค แอซิดซึ่งจะสามารถช่วยลดขั้นตอนการปรับยาในผู้ป่วยได้*

คำสำคัญ : *วาลโปรอิค แอซิด, พารามิเตอร์ทางเภสัชจลนศาสตร์, กลุ่มผู้ป่วยควบคุมอาการชักได้, กลุ่มผู้ป่วยควบคุมอาการชักไม่ได้.*

Epilepsy is one of the most common neurological problems. Its annual incidence ranges from 20 to 70 cases per 100,000 populations.[1] In Thailand, the total prevalence of epilepsy in the general population whose ages were more than five years in the years 1991-1992 was 29.2 cases per 1,000 populations and the prevalence of active epilepsy was 5.9 cases per 1,000 populations.[2] Most people who develop epilepsy have a relatively short-lasting susceptibility to seizures and enter their remission shortly after starting treatment on small doses of antiepileptic drugs (AEDs).[1, 3] However, some patients do not become completely free of seizure; 20 to 30 percent of the people who develop epilepsy have chronic epilepsy that responds that incompletely responds to AED therapy, despite the choice of an adequate AED and carefully monitored treatment.[4] It is not known why and how epilepsy becomes drug resistant in some patients while others with seemingly identical seizure types and epilepsy syndromes can achieve seizure control with medication. The consequences of drug-resistant epilepsy can be quite severe, including mortality rates that are four to seven times higher in those with drug-resistant seizures.[5] Factors associated with seizure control were symptomatic etiology, combination of symptomatic etiology with mental retardation, long duration of epilepsy, partial epilepsy and cognitive deficits.[6] Knowledge of pharmacokinetics (PK) of each antiepileptic drug will lead to better antiepileptic drug concentration monitoring and will improve outcome.

Valproic acid (VPA) is a branched-chain fatty acid. The drug and its derivatives have been available for use as antiepileptic drugs since 1960s.[7] It is currently one of the primary drug used to treat various seizure disorders. VPA is largely bound to plasma proteins and has a relatively small Vd. It is 90-95 percent bound to albumin, the binding sites for VPA are saturable at therapeutic plasma levels, and the free fraction may increase as the total concentration increase. The half life (t $\frac{1}{2}$) of VPA varies from 10 to 20 hours in adults while it is significantly shorter in children.[8]

Fernando-dongas M.C. et al. studied the characteristics of VPA-resistant juvenile myoclonic epilepsy (JME). They reported that the VPA-resistant group had a higher incidence of atypical characteristics including asymmetric EEG abnormalities, atypical seizure characteristics and history, and neurological and neuroimaging abnormalities.[9] VPA-resistant JME have higher incidence of focal EEG abnormalities which is suggestive of partial epilepsy. This raises a question: whether the VPA- resistant JME group is in fact refractory localization related epilepsy. Shen D.D. et al. studied the distribution of VPA between the brain (gray matter) and serum in patients with intractable seizures who were on chronic VPA therapy. There was a tendency for the brain-to-serum concentration ratio to be lower in tissues from the epileptic foci than in tissues from non-epileptic areas.[10] This raises another question: whether the characteristics of the brains of epileptic patients who should or should not respond to VPA are associated with their VPA PK values. Panomvana D. et al. had found that the volume of distribution (Vd) of VPA was significantly narrower while the half-life (t $\frac{1}{2}$) was significantly shorter in VPA-resistant pediatric patients than in VPA-responsive pediatric patients. The same results were

obtained for both total and unbound VPA.[11]

This study was performed to determine whether there is any difference of VPA PK parameter values of the patients and their responsiveness to VPA therapy, and whether or not the estimated PK parameter values of the patient could be used to distinguish between VPA-resistant and VPA-responsive adult epileptic patients.

Methods

Patients

The subjects included in this study were epileptic patients who were 15-65 years old who came for their follow up at the Neurology Clinic, Neurology Institute. The study protocol has been approved by the ethic committee of the Neurology Institute. VPA-responsive patients were the patients who had no seizures since start taking VPA monotherapy or the frequency of seizure had decreased more than 50 % as compared to before their taking VPA. VPA-resistant patients were the patients who were treated with VPA monotherapy in appropriate dose with good compliance for a period of time (not less than four weeks), and the frequency of seizure was not decreased or was decreased less than 50 % as compared to before taking VPA, or other antiepileptic drug had to be added into the therapy.

Inclusion Criteria

Patients who were diagnosed as having epilepsy with their age 15-65 years old in either sex, they were treated with VPA (Depakine Chrono® 500 mg) either monotherapy or polytherapy which did not include phenobarbital, phenytoin, carbamazepine or other drugs that use the same metabolic enzyme pathway as VPA.

Exclusion Criteria

Patients who had history of VPA allergy, clinical sign of abnormal in hepatic or renal functions, precipitating factor for seizures, pregnancy and lactation, poor or noncompliance.

Blood Samples Collection

All patients consumed Depakine Chrono® 500 mg dosage form. The dosing interval was modified from either *tid* or *bid* to once daily regimen at least seven days prior to blood sample collection to assure steady state condition. Two blood samples were collected at least 14 hours after the intake of the studied dose to ensure elimination phase. They were collected approximately five hours apart, and the exact times were recorded.

Drug Analysis

Each blood sample was allowed to clot and was centrifuged immediately at 5,000 rpm for seven minutes at room temperature. The serum was then separated and frozen at -20 c^o until the time of assay. Serum levels of total and unbound VPA were measured by Fluorescence polarization immunoassay (TDX FLx Abbott laboratories)[12]; unbound VPA levels were determined after ultrafiltration (25 ^{o}C, 2000 g, 35 fixed angle x 20 min, Centrifree TM Micropartition System).[13] Serum albumin level was also measured from the same serum sample.

Data Analysis

1. Pharmacokinetic Data Analysis

Individual VPA PK parameter values were estimated using the equations for a linear one-

compartment-model with a first-order declining of drug concentration after the end of 14 hours zero-order (constant rate) drug release from the intake of Depakine Chrono dosage form. The equations used were:

$$k = \frac{[\ln c_{t1}/c_{t2}]}{\Delta t}$$

$$t{:}\tfrac{1}{2} = \frac{0.693}{k}$$

$$Vd = \frac{[SFD/t_{inf}]}{k\,C_t}\ \frac{[1-e^{-ktinf}]}{1-e^{-kt}}\ e^{-k(t-tinf)}$$

$$Cl = k \times Vd$$

C_t : Drug concentration at time t

F : Bioavailability

D : Dose

V_d : Volume of distribution

t_{inf} : Infusion time equal 14 hour

τ : Dosing interval

k : Elimination rate constant

Cl: Clearance

$t{:}\tfrac{1}{2}$ Half-life

S : Fraction of active drug accounting for salt form

2. Statistical Analysis

Demographic data were analyzed and presented as percentage and mean ± SD, the values of the two groups were compared using either *chi*-square or Student's t-test The PK parameter values of both total and unbound VPA of the patients in either VPA- responsive or VPA-resistant groups were presented as mean ± SD. PK parameters of the two groups were compared by Mann-Whitney test. The associations between VPA PK parameter values and the responsiveness to VPA therapy were also determined using *chi*-square test and the odds ratio.

Results

Thirty-three patients were enrolled into the study. There were 19 patients (57.6%) who had no seizures after treatment with VPA and fourteen patients (42.4%) who still had seizures even after high dose of VPA was administered.

1. Demographic Data

Table 1 shows that there were no statistically significant differences in sex, age, weight, and albumin levels between the patients in VPA-responsive and VPA-resistant groups while the dose/day was of borderline significantly higher in the VPA- resistant group. The seizure types of the two groups were significantly different; majority of patients in the VPA-responsive group had generalized tonic clonic seizure where as in the VPA-resistant group had complex partial seizure. Table 2 shows comparisons of the percentage of abnormality found by EEG, MRI and

CT between the seizure-controlled and seizure-uncontrolled by VPA groups. There were no statistically significant differences in the frequency of abnormality found by EEG and CT between the two groups. However, the MRI tended to detect higher incidence of abnormality in the VPA non-responsive group at the significant level of $p < 0.1$. Five patients in the VPA-resistant group had weight gain and one patient had tremor. Nine patients in the VPA-responsive group had weight gain while two patients had tremor.

Table 1. Demographic data.

Characteristic	VPA-resistant group (n=14)	VPA-responsive group (n=19)	*p*-value
Sex, n (%)			
- Male	7 (50.0)	5 (20.0)	0.162[a]
- Female	7 (50.0)	14 (80.0)	
Age (years), mean ± SD (range)	30.2629 ± 10.58085 (16.02-49.03)	33.7311 ± 12.0624 (15.03 - 56.05)	0.397[b]
Weight (kg), mean ± SD (range)	63.4857 ± 13.07816 (43.50 - 88.00)	59.1105 ± 11.2617 (46.00 - 88.00)	0.311[b]
Dose/day (mg/kg), mean ± SD (range)	20.6457 ± 5.96969 (10.07 - 33.98)	17.5400 ± 2.9289 (10.87 - 21.93)	0.057[b]
Albumin (g/dL), mean ± SD (range)	3.8429 ± 0.45694 (3.10 - 4.60)	3.9053 ± 0.2697 (3.50 - 4.70)	0.626[b]
Seizure types[c], n (%)			
Partial seizure			
1. Simple partial seizure	1 (7.20)	1 (5.26)	
2. Complex partial seizure	7 (50.00)	3 (15.80)	
3. Complex partial seizure with generalized tonic- clonic seizures	3 (21.40)	1 (5.26)	0.027[a]
Generalized seizures			
1. Generalized tonic- clonic seizures	3 (21.40)	14 (73.68)	

[a]Chi-square test

[b]Student t-test

[c] Based on International League Against Epilepsy 1981

Table 2. Neurological examinations.

Neurological examination	VPA-resistant group (n=14)		VPA-responsive group (n=19)		p-value[a]
	Total n	Abnormal n (%)	Total n	Abnormal n (%)	
EEG	9	7 (77.78)	10	9 (90.00)	0.582
MRI	7	6 (85.71)	4	1 (25.00)	0.088
CT	4	1 (25.00)	7	3 (42.86)	1.000

Total n = Total number of patients examined by the device

[a] *Chi*-square test

2. Therapeutic Concentrations

Total VPA concentrations determined from blood samples collected from the patients either in the VPA-responsive or VPA- resistant groups were mostly within the proposed therapeutic range (50 - 100 mg/L), several patients showed total measured VPA concentrations at 14th hour after drug administration to be higher than 100 mg/L while a few patients showed total VPA concentrations at 24th hour after drug administration to be less than 50 mg/L. High percentage of patients in the seizure-uncontrolled group had total VPA concentrations within or higher than the proposed therapeutic range, but still the seizures could not be controlled. Lower percentage of patients in the seizure-controlled group had their total VPA concentrations within the proposed therapeutic range as compared to the seizure-uncontrolled group. A few patients in this group had their total VPA concentrations at 24th hour after drug administration which were less than 50 mg/L but their seizures were under controlled.

3. Pharmacokinetic Parameters

Comparisons of the PK parameters of VPA; elimination rate constant (k), half-life (t ½), volume of distribution (Vd) and clearance (Cl) between VPA-resistant and VPA-responsive groups are shown in table 3 and table 4.

For total VPA concentrations, the mean/median value of Vd of the VPA-resistant group was less extensive and the $t_{\frac{1}{2}}$ was shorter when compared to the VPA- responsive group with p value equaled to 0.018 and 0.069 respectively. Chi-square test also showed that there was significant association between Vd values with the responsiveness of the patients to VPA treatment at $p < 0.05$. Patient who had total VPA Vd that was less than 0.3 l/kg had a higher tendency to belong to the VPA resistant group at significant level $p < 0.05$. While, k and $t_{\frac{1}{2}}$ that was shorter than 25 hours of total VPA tended to associate with the responsiveness of the patients to VPA treatment but they did not reach statistical significance. No significant difference in Cl of the total VPA between the two groups could be observed.

For unbound VPA concentrations, comparisons of the PK parameters obtained also indicated that the mean/median value of $t_{\frac{1}{2}}$ in the VPA- resistant group was less than the value in the VPA-responsive group while the Vd was smaller in the

Table 3. Comparisons of the mean/median pharmacokinetic parameter values between VPA-resistant and VPA responsive groups.

Pharmacokinetic parameters	VPA-resistant group (n = 14)		VPA-responsive group (n = 19)		p value[a]
	Mean ± SD	Median	Mean ± SD	Median	
Total VPA concentrations					
K (hr^{-1})	0.0394 ± 0.0123	0.0358	0.0307 ± 0.0149	0.0275	0.069
t $^{1}/_{2}$ (hr)	19.1088 ± 5.4373	19.3710	29.0132 ± 16.3237	25.1945	0.069
Vd (L/kg)	0.2209 ± 0.0764	0.2149	0.3073 ± 0.1084	0.2988	0.018
Cl (L/hr/kg)	0.0083 ± 0.0031	0.0080	0.0081 ± 0.0025	0.0075	0.870
Unbound VPA concentrations					
K (hr^{-1})	0.0703 ± 0.0235	0.0724	0.0555 ± 0.03167	0.0578	0.109
t $^{1}/_{2}$ (hr)	11.2232 ± 4.8796	9.6690	35.2704 ± 72.4589	11.9905	0.109
Vd (L/kg)	1.1370 ± 0.9614	0.6253	3.3902 ± 6.3914	1.0845	0.190
Cl (L/hr/kg)	0.0690 ± 0.0539	0.0473	0.0709 ± 0.0517	0.0587	0.560

[a] Mann-Whitney test

Table 4. Association between pharmacokinetics parameter values and the responsiveness to valproic acid therapy.

PK parameters of VPA Factor present	VPA resistant group		VPA responsive group		p-value	Odds ratio	95%CI	
	Y	N	Y	N			Lower	Upper
Total VPA								
K ≥ 0.03 hr^{-1}	11	3	9	10	0.070[a]	4.074	0.854	19.432
t $^{1}/_{2}$ ≤ 25 hr	11	3	9	10	0.070[a]	4.074	0.854	19.432
Vd ≤ 0.30 l/kg	10	4	7	12	0.049[a**]	4.286	0.968	18.973
Cl ≥ 0.009 l/kg/hr	7	7	9	10	0.851[a]	1.111	0.279	4.423
Unbound VPA								
K ≥ 0.07 hr^{-1}	7	7	4	15	0.136[b]	3.750	0.819	17.166
t $^{1}/_{2}$ ≤ 10 hr	7	7	4	15	0.136[b]	3.750	0.819	17.166
Vd ≤ 0.70 l/kg	7	7	3	16	0.057[b]	5.333	1.058	26.898
Cl ≥ 0.05 l/kg/hr	8	6	16	3	0.122[b]	0.250	0.049	1.270

Y= yes; N= no

[a] *Chi*-square test

[b] Fischer's exact test

** Statistical significant at p < 0.05

VPA non-responsive group as compared to the VPA-responsive group. However, these differences were not significant at p=0.05 but were significant at p = 0.109 and 0.190 (Mann-Whitney two-sided test), respectively. Additional tests using Fischer's exact test and the odds ratio indicated that Vd of unbound VPA which was less than 0.7 l/kg and t ½ which was less than 10 hours had a higher probability to belong to the VPA-resistant group at significant level of p < 0.1. Cl of the unbound VPA showed no significant difference between the two groups.

Discussion

1. Demographic Data

The data were obtained from 33 adult patients using VPA. Nineteen patients were in the VPA-responsive group; these patients were taking VPA as a monotherapy. On the other hand, 14 patients in VPA-resistant group were either taking VPA monotherapy or added on some new generation antiepileptic drugs, such as topiramate, lamotrigine, gabapantin, oxcarbazepine and levetiracetam. The dose/day was barely significantly higher in the VPA-resistant group indicated that adequate dosage had been given to the patient in the VPA-resistant group and further increase in the dosage of VPA would not be the answer to the uncontrolled seizures especially that the serum VPA concentrations in general were also seemingly higher.

The seizure types were significantly different between the two groups; the most common seizure type in the VPA-resistant group was complex partial seizures (50.00 %), this was in agreement with previous study which indicated that about 60 % of patients with intractable epilepsy suffer from partial

seizures.[3] The most common seizure type in the VPA-responsive group was generalized tonic clonic seizures (73.68 %) which were consistent with a previous study which reported that VPA treatment in 808 adults and 585 children with epilepsy resulted in more than 75% reduction in seizure frequency in 78% of patients with generalized seizures.[3] Most patients had their albumin values within the normal ranges. There were no statistically significant differences in percentages of neurological abnormality detected from EEG and CT examinations between VPA responsive and VPA resistant groups, these indicated that the abnormality detected by the two devices could not be used as an indicator for seizure controlled or not controlled by VPA. However, regarding the MRI, the difference in percentage of abnormality found between the two groups was significant at p=0.088, VPA-resistant patients had higher tendency to show abnormality from MRI. This result was consistent with Fernando-dongas M.C. et al. who suggested that the VPA-resistant group had a higher incidence of neurological and neuroimaging abnormalities.[9]

2. Pharmacokinetic Parameter Values

The PK parameter values obtained were highly varied among patients. Several patients showed extreme values. Only two blood samples only were collected. They were taken at least 14 hours after drug intake, based on a previous report that total and unbound plasma concentrations of VPA would be stable at plateau from 4-14 hours after administration of Depakine Chrono dosage form[14] which implies that after 14th hour, the absorption process should fade out and the elimination phase would dominate. However, it is quite possible that, in

some patient, the absorption process is still continuing after 14th hour and the elimination phase is not yet approached. We tried to cover this uncertainty by collecting the two blood samples as late as possible in our later cases to ensure the elimination phase. The two blood samples of several patients were collected at 24th and 29th hours after drug intake and for a few patients, the two blood samples were collected as late as 36th and 41st hours after drug intake. No significant difference in the elimination rate constant or half-life could be observed between the values obtained from the blood samples collected in the earlier or later cases.

The $t\frac{1}{2}$ of VPA in VPA-resistant group was shorter and the Vd was less extensive than those obtained in VPA-responsive group. These might imply that less VPA could distribute into the brain of the seizure-uncontrolled by VPA group. The uptake and exit of VPA within the central nervous system can occur at the choroids plexus of the blood brain barrier. Approximately two-third of the VPA uptakes occurs through a saturable carrier-mediated process and a lesser degree through passive diffusion. A relatively small amount of VPA is distributed by passive diffusion into tissues, as only the non-ionized, i.e., lipid soluble portion of VPA diffuses across membranes. VPA has a pKa of 4.56 and thus at physiological pH 7.42, VPA is highly ionized into a carboxylate moiety, valproate. [15] The apparent Vd of VPA is relatively small, it is variable and usually ranges from 0.1 to 0.5 l/kg.[8] The drug reaches the brain in concentrations ranging from 7-28 % of the simultaneous concentration in plasma, while CSF levels are 8-25 % of those in plasma.[8] The CSF levels were lower than plasma and subcutaneous ECF levels, particularly at

steady state. This suggests that VPA may be a substrate for an energy-dependent carrier-mediated transport in and out of the CNS. The reason for having low Vd in the VPA non-responsive group is not fully understood. There might be some mechanism in the blood brain barrier that decreases the movement of VPA into CNS or the brain uptake process might become saturated more easily in some patients. VPA might not bind to lipid or protein components of neural tissues.[10, 16] Lower Vd means higher VPA concentrations in the plasma resulting in higher proportion of the drug which can distribute to the elimination organ, and in turn, higher k and shorter $t\frac{1}{2}$ of VPA were obtained in the VPA non-responsive group.

The Vd values estimated from the total concentrations obtained in our study were within the range of 0.1-0.5 l/kg as previously reported.[8] The lower end of $t\frac{1}{2}$ was closed to 10 hours as previously reported. However, the upper end was much higher than those reported earlier, partly might due to the multiple- dose conditions and the chrono dosage form consumed in this study. The Cl values were closed to the values reported by Cavanaugh et al. who reported the Cl of VPA obtained from total VPA concentrations after consuming a controlled-release formulation, under multiple-dose conditions to be 8.37 ml/hr/kg.[17] Our Cl values were also comparable with those obtained in a recent study by Dutta et al. who reported the mean Cl (standard deviation) values for total and unbound VPA in adolescents after once-daily administration of divalproex sodium extended-release tablets to be 9.06 (2.03) and 82.3 (28.2) ml/hr/kg, respectively. [16]

Since the sample size was small and several outlier values were obtained, the Mann-Whitney test which is a test for comparison of the median values was chosen in place of the test for the comparison of the mean values. *Chi*-square test and the odds ratio were also performed to set up the PK parameter limited values obtained from total VPA between the two groups. Fischer's exact test instead of *chi*-square test was used to compare PK parameters obtained from unbound VPA due to these data violated the assumption of *chi*-square testing.

There were several limitations in this study; the PK parameter values were obtained from two blood samples only, error either in any of the sampling time or drug concentration assay could lead to significant error in the PK parameter value estimated. The times when blood samples were collected, might not yet approach the elimination phase due to the sustained-release dosage form consumed. Many confounding factors were involved, resulting in high variation of the PK parameter value estimated even among patients within the same group. The number of patients included into the study was quite small while the standard deviation was large for each PK parameter. The significant levels found were therefore mostly at the < 0.1 level. Only the Vd obtained from total VPA was found statistically significantly different at $p < 0.05$.

Conclusion

Vd of VPA were significantly different between patients in the VPA-responsive and VPA-resistant groups. A narrow Vd could be an indicator that the seizure of the patient had high tendency of could not be controlled by VPA. Besides, $t_{1/2}$ of VPA tended to be shorter in the VPA non-responsive group as compared to the VPA responsive group.

Two blood samples collected during the elimination phase of VPA in the patient, besides being used for routine therapeutic drug level monitoring to relate the VPA concentrations obtained with efficacy and adverse drug reaction as normally performed in clinical practice. The corresponding estimated PK parameters might be used for identifying whether or not the epileptic patients will response favorably to VPA. A seizure-uncontrolled by VPA patient has a higher incidence of neurological and neuroimaging abnormalities by MRI. However, the MRI examination is quite expensive, if the VPA concentrations obtained from routine clinical therapeutic drug levels monitoring could help to distinguish between the two groups of patients, this could be helpful; the steps of trial and error with the unresponsive drug could then be reduced.

Further studies with higher number of subjects and in more conservative designs are required before any definite conclusion can be made.

Acknowledgements

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