PHARMACOTHERAPY

BRIEF REPORT

Valproate Protein Binding Is Highly Variable in ICU Patients and Not Predicted by Total Serum Concentrations: A Case Series and Literature Review

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Study Objective The free fraction of valproate (the pharmacologically active moiety, normally 5–10%) may vary significantly in critically ill patients, but this topic is understudied, with only four prior intensive care unit (ICU) case reports. The objective of this study was to evaluate the range of valproate plasma protein binding in ICU patients.

Design Observational study of consecutive ICU patients.

Setting Neurocritical and medical critical care services in a nonuniversity academic medical center.

Patients Consecutive ICU patients treated with valproate with serum albumin less than 4 g/dl.

Measurements and Main Results Simultaneous total and free trough serum valproate concentrations were measured as were serum creatinine, blood urea nitrogen, albumin, platelets, and transaminase values. The reference concentration range was 50–125 mg/L (total) and 5–17 mg/L (free). Valproate concentrations were categorized as within reference range, low, or high, and as concordant if both concentrations were in the same category. Data are reported as median (interquartile range). Fifteen patients (nine men) were evaluated. The median age was 63 (34–70) years. The valproate dose was 3 g/day (35 mg/kg/day). No patient had a valproate free fraction of 5–10%; the median was 48%, and the range was 15–89%. Total and free concentrations showed poor correlation (0.43) and were concordant in only two patients (both in the reference range). Free valproate concentration was poorly predicted by an equation correcting for albumin ($r = 0.45$). Suspected adverse drug events occurred in 10 patients: hyperammonemia in 7 of 12 tested (58%), elevated transaminases in 2 of 15 (13%), and thrombocytopenia in 5 of 15 (33%).

Conclusions Protein binding of valproate was highly inconsistent in this cohort of ICU patients, and total valproate concentrations did not predict free concentrations, even when correcting for albumin. Additional research to define best practice for dosing and monitoring valproate and the relationship between free valproate concentrations and clinical or adverse effects in ICU patients is needed.

Key Words valproate, valproic acid, divalproex, adverse drug effects, myoclonus, protein binding, therapeutic drug monitoring, seizure, intensive care.

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Conflict of Interest: The authors declare that they have no conflict of interest.

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Valproic acid is an antiepileptic and mood-stabilizing agent approved by the Food and Drug Administration in 1978, and the injectable valproate was approved in 1996. Doses are typically adjusted to a total serum valproate concentration of 50–100 mg/L when treating seizures and 50–125 mg/L for behavioral disorders.[1, 2] Valproate is highly protein-bound to albumin (90% or higher), and the pharmacologically active unbound or free fraction is typically 5–10% of the total valproate concentration but may vary significantly.[3] Valproate protein binding has been understudied; we are aware of only four reported cases of free valproate monitoring in intensive care unit (ICU) patients.[4–7] Reviews and guidelines recommend monitoring serum valproate concentrations but make sparse or no mention of free serum concentration monitoring or altered protein binding.[8–12]

Our interest in monitoring free serum valproate concentrations began after treating a patient with decreasing total serum valproate concentrations despite increasing doses, with several adverse drug events. The free fraction and free serum valproate concentrations were elevated despite a low total concentration less than 30 mg/L. The primary objective of this study was to evaluate the frequency and extent of deranged valproate plasma protein binding in a cohort of consecutive critically ill patients. The secondary objective was to assess the capability of a previously published equation to predict free serum valproate concentrations from albumin and total valproate concentrations.[13, 14]

Methods

Index Case

A 72-year-old man weighing 103 kg remained comatose after in-hospital cardiac arrest. Targeted temperature management was initiated at 33°C for 24 hours, with controlled rewarming over 12 hours. On hospital day (HD) 3, he developed intermittent myoclonus with generalized polyspike and wave discharges of high amplitude on electroencephalography that persisted despite levetiracetam and continuous midazolam therapy. Because neuron-specific enolase testing and magnetic resonance imaging on HD4 did not identify severe brain injury, aggressive care was continued. Valproate (30 mg/kg/day) and a propofol infusion were added on HD4. On HD7, the total serum valproate concentration was 71 mg/L (reference range 50–100 mg/L), but a repeat total concentration on HD12 was low (31 mg/L) despite no change in valproate dose (Figure 1). A 2000-mg dose of valproate was administered, and the maintenance dose increased to 40 mg/kg/day.

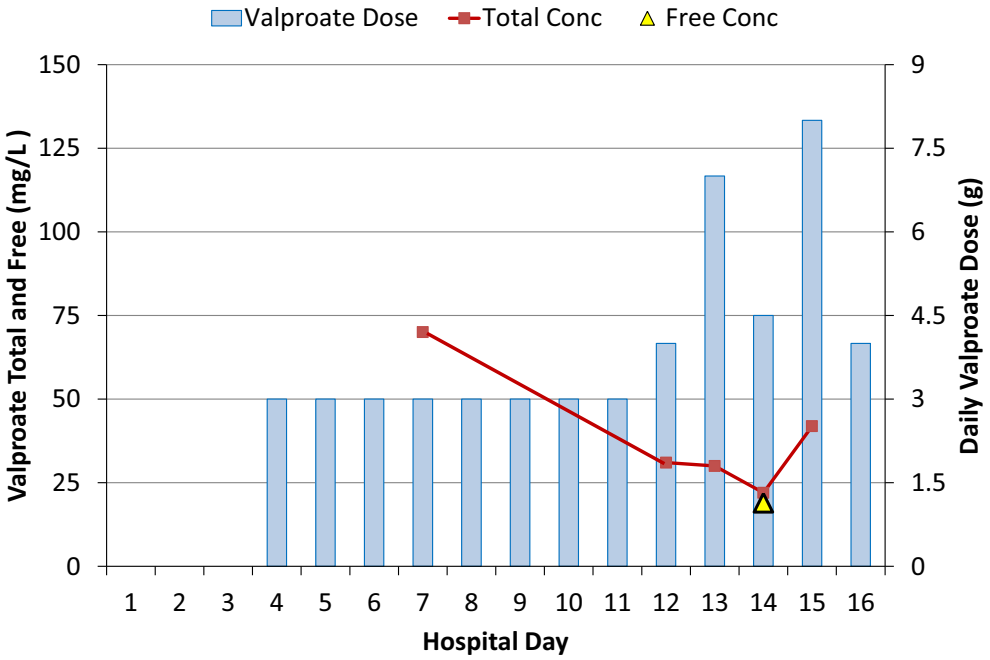

Figure 1. Index case data showing decreasing total serum valproate concentrations (squares) despite repeated dose increases (vertical bars), and a free fraction on day 14 of 86% (free concentration 19 mg/L; large triangle, total concentration 22 mg/L).

On HD13, the total concentration was 30 mg/L, again prompting a valproate bolus and increase in the maintenance regimen to 60 mg/kg/day. On HD14, the total concentration was lower at 26 mg/L, the serum albumin was 2.2 g/dl, and the dose increased again to 80 mg/kg/day. Serum for total and free valproate concentration measurement was sent to a reference laboratory, which returned on HD16, revealing an even lower total concentration (22 mg/L), an elevated free concentration of 19 mg/L (reference range 5–17 mg/L), and a dramatically elevated free fraction of 86%. Coupled with an elevated ammonia concentration (135 μmol/L) and worsening transaminases, valproate therapy was discontinued. After repeated failures to control epileptiform discharges, the family requested withdrawal of life-sustaining therapy.

Study Design

Maine Medical Center is an academic medical center with 58 adult ICU beds. We prospectively monitored consecutive adult patients admitted to the neurocritical or medical critical care services from September 2015 through August 2016 who were treated with valproate with serum albumin concentrations lower than 4.0 g/dl. When the bedside treatment team ordered valproate concentrations, simultaneous total and free serum valproate concentrations were obtained. Total and free valproate assays were sent to a reference laboratory (Mayo Clinic Medical Laboratories, Rochester, MN) with a delay of several days. Total and free serum valproate concentrations were determined via enzyme immunoassay using the Roche Cobas 8000 system. Free serum valproate was measured after ultrafiltration with the Millipore Centrifree micropartition system (EMD Millipore, Billerica, MA). In response to the index case, this study was designed as a Quality Improvement pilot project with approval by our institutional review board.

We collected patient demographic data including age, gender, weight, reason for ICU admission, length of stay, serum albumin, blood urea nitrogen, and creatinine concentration on the day simultaneous free and total serum concentrations were measured. If more than one free valproate concentration was obtained, the sample with the highest free fraction was used.

Drug administration data were collected from the electronic Medication Administration Record, including the dose (g/day and mg/kg/day) on the day the serum concentrations were obtained, indication for valproate therapy, whether valproate was started during this ICU admission, and time from valproate initiation to measurement of serum concentrations. Concomitant administration of aspirin, propofol, and/or Intralipid (Baxter Healthcare Corp., Franklin Lakes, NJ) was recorded. Inappropriate dose escalation was defined as an increase in valproate dose based on total serum valproate concentration when the free concentration suggested the dose should not have been increased.

Outcomes and Statistical Analysis

We assessed possible valproate-induced adverse effects during valproate administration until hospital discharge. Only patients who had baseline laboratory values prior to valproate initiation were assessed for laboratory-based adverse effects. Hepatotoxicity was defined as a new alanine aminotransferase (ALT) more than 3 times the upper limit of normal (ULN; more than 120 U/L), alkaline phosphatase more than 2 times the ULN (more than 234 U/L), total bilirubin more than 2 times the ULN (more than 2 mg/dl), or a doubling of the baseline value for ALT, alkaline phosphatase, or bilirubin if abnormal prior to valproate initiation.[15] Thrombocytopenia was defined as a platelet count lower than 140,000 cells/mm^3 or reduction in platelet count by more than 50% if platelets were already lower than 140,000 cells/mm^3 following valproate initiation.[16] Hyperammonemia was defined as a new serum ammonia level of 60 μmol/L or more following valproate initiation.

We compared the measured free serum valproate concentration with a predicted free serum concentration derived from this equation: free fraction (%) = Ae^{-BX}, where X corresponds to albumin concentration (mmol/L), with constants $A = 130.69$ and $B = 4.96 \times 10^{-3}$.[13, 14] The total, free, and predicted free serum valproate concentrations were compared using the Spearman correlation, and the bias between mean differences was assessed with Altman-Bland analysis by Analyse-it software.[17] Concentrations were categorized relative to the reference ranges for total (50–125 mg/L) and free (5–17 mg/L) serum valproate concentration as in the reference range or below ("low") or above the reference range ("high"). If both total and free concentrations were in the same category, we considered them therapeutically concordant. If

the concentrations were in different categories, they were considered therapeutically discordant. Data are presented as median (interquartile range [IQR]) with $p<0.05$ considered significant.

Results

From September 2015 through August 2016, 15 medical or neurologic critical care patients were monitored (including the index case). The subjects were mostly male with a wide distribution of ages (Table 1). Valproate was administered for seizures (six patients), myoclonus (four patients), bipolar disorder (three patients), schizophrenia (one patient), or refractory agitated behavior (one patient). Three patients (20%) were receiving valproate at the time of ICU admission at the same dose used prior to hospitalization.

The total serum valproate concentration was 52 mg/L (IQR 29–65 mg/L) with a range of 22–92 mg/L, representing low or reference range values only. No patient had a valproate free fraction of 5–10%; the median unbound or free valproate fraction was 48% (IQR 39–56%; range 15–89%). Despite the low or reference range total valproate levels, the increased free fraction was associated with free serum valproate concentrations that were often above the reference range at 20 mg/L (IQR 18–30 mg/L; range 11–37 mg/L). The median valproate dose was 3.0 g/day (IQR 2.0–4.0 g/day) or 35 mg/kg/day (IQR 27–43 mg/kg/day). On the day valproate concentrations were drawn (drug day 4 [IQR 3–11]), the median albumin value was 2.4 g/dl (IQR 2.2–3.0 g/dl), blood urea nitrogen was 21 mg/dl (IQR 15–28 mg/dl), and serum creatinine was 0.87 mg/dl (IQR 0.56–1.2 mg/dl). Eight patients (53%) were receiving medications known to alter valproate protein binding: propofol infusions (five patients), aspirin (two patients), and both propofol and aspirin (one patient; Table 2). The median time from drawing the serum sample to valproate concentration availability was 3 days (range 2–4 days).

Table 1. Patient Demographics and Clinical Outcomes

Demographics	
Age, yr	63 (29–72)
Male	9 (60)
Ethnicity, white	14 (93)
ICU admitting diagnosis	
Cardiac arrest	5 (33)
Seizures	4 (27)
Acute respiratory failure	4 (27)
Sepsis	1 (7)
HSV encephalitis	1 (7)
Weight, kg, median (IQR)	95 (73–109)
Outcomes	
ICU mortality	3 (20)
Hospital mortality	6 (40)
Discharge disposition	
Deceased	6 (40)
Rehabilitation	4 (27)
Home	3 (20)
Transfer	2 (13)
ICU length of stay, days, median (IQR)	11 (8–27)
Hospital length of stay, days	21 (10–34)

HSV = herpes simplex virus; ICU = intensive care unit; IQR = interquartile range.
Continuous variables reported as median (IQR), nominal variables as frequency n (%).

The total serum valproate concentration was low in six patients (40%) and in reference range in nine (60%; Figure 2). The free valproate concentration was never low, but was high in 12 patients (80%) and in reference range in 3 (20%). Total and free concentrations were concordant in only two patients (13%). Among the six patients with low total concentrations, which would likely have resulted in valproate dose increases based on the total concentration, free concentrations were high in five (83%).

The correlation was poor between total and free serum valproate concentration ($r = 0.43$), between measured and predicted free serum valproate concentration ($r = 0.45$; Figure 3), and between albumin and free fraction ($r = 0.21$). When the bias between measured and estimated-free serum valproate concentrations using the Parent formula was compared using Altman-Bland techniques (Figure S1), the Parent formula consistently underestimated free concentrations with a bias of −11.9 mg/L and 95% limits of agreement of −28 to 4.2.

Adverse drug events possibly associated with valproate therapy occurred in 10 patients (68%; Figure 2). Seven (58%) of 12 patients tested had hyperammonemia, 2 (13%) of 15 developed elevated transaminases, and 5 (33%) of 15 had thrombocytopenia. Unexpectedly, four of the five patients without adverse drug events had among the highest free concentrations recorded in this study. The first two patients monitored received inappropriate dose increases based on low total serum valproate concentrations. Learning from those events, providers checked free serum valproate concentrations earlier, and none of the next 13 patients received dose supplements inappropriately.

Discussion

No patient in our consecutive series had a normal free fraction of valproate of 5–10%;

Table 2. Prospective 15 ICU Cases of Free Valproate Monitoring and 4 Previously Reported Cases

Case	Age, yrs	Gender	Dose, g	Albumin, g/dl	BUN, mg/dl	Creatinine, mg/dl	Drugs[a]	Total valproate, mg/L	Free valproate, mg/L	Free fraction	Adverse events
1	72	M	6	2.2	27	0.87	P, S	22.2	19	0.86	A, H, D
2	62	M	4	2	49	1.14	–	53	20	0.38	A, D
3	93	F	3	2.4	27	1.06	P	71	37	0.52	–
4	69	M	2	2.7	8	0.56	–	53	32	0.6	–
5	19	F	4	3	9	0.32	–	42	18	0.43	A
6	72	F	1	2.1	57	3.3	S	52.1	18	0.35	A, T
7	26	M	3	2.4	18	0.57	P	29.1	26	0.89	A, T
8	69	M	3	2.4	33	1.21	–	65	31	0.48	–
9	67	F	2	3.2	26	5.6	–	23	19	0.83	–
10	81	M	2.25	2.2	8	0.57	P	73	11	0.15	T
11	51	F	3.6	2.7	12	0.41	–	47	22	0.47	A
12	24	M	4	2.9	29	4.3	P	27	13	0.48	H, T
13	29	F	1	3.7	19	0.95	–	51	12	0.24	A
14	63	M	4.5	3	19	0.49	S	56	29	0.52	–
15	40	M	3	2.2	21	0.57	P	92	37	0.4	T
Median (IQR) or N (%)	63 (34–70)	9 M (60%)	3 (2–4)	2.4 (2.2–3.0)	21 (15–28)	0.87 (0.56–1.2)	6 P (40%) 3 S (20%)	52 (29–65)	20 (18–30)	0.48 (0.39–0.56)	7A, 2 H, 5 T, 2 D
Ref 4	52	F	3.5	1.6	[b]	[b]	[b]	46	35	0.76	T, D
Ref 5	29	[b]	[b]	0.8	[b]	[b]	–	14	13	0.93	D
Ref 6	61	F	0.9	1.2	[b]	[b]	–	15.5	9.3	0.60	D
Ref 7	53	M	2.4	2.1	[b]	[b]	[b]	62	17	0.27	A

A = ammonia elevation; BUN = blood urea nitrogen; D = drug dosing error; F = female; H = hepatic dysfunction; M = male; P = propofol, S = salicylate; T = thrombocytopenia.
Data are presented as incidence (%) and median (interquartile range).
[a]Drugs = medications that alter valproate protein binding.
[b]Data not reported.

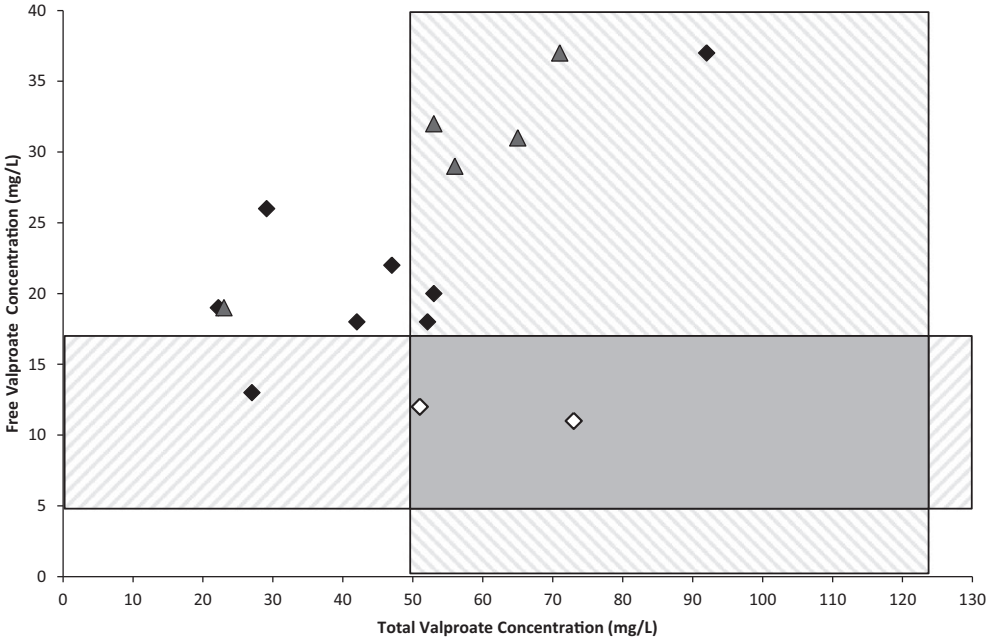

Figure 2. Simultaneous free and total valproate concentrations. The diagonal up to right shading reflects the reference range for the free concentration on the y-axis (5–17 mg/L), the diagonal up to the left shading reflects the reference range for the total concentration on the x-axis (50–125 mg/L), and the solid gray rectangle reflects the concordance of these two reference ranges. Triangles represent patients without adverse events. Diamonds represent patients with adverse events, two of which were therapeutically concordant (both in reference range with unfilled diamonds). The remaining 13 symbols are solid diamonds or triangles reflecting therapeutically discordant values.

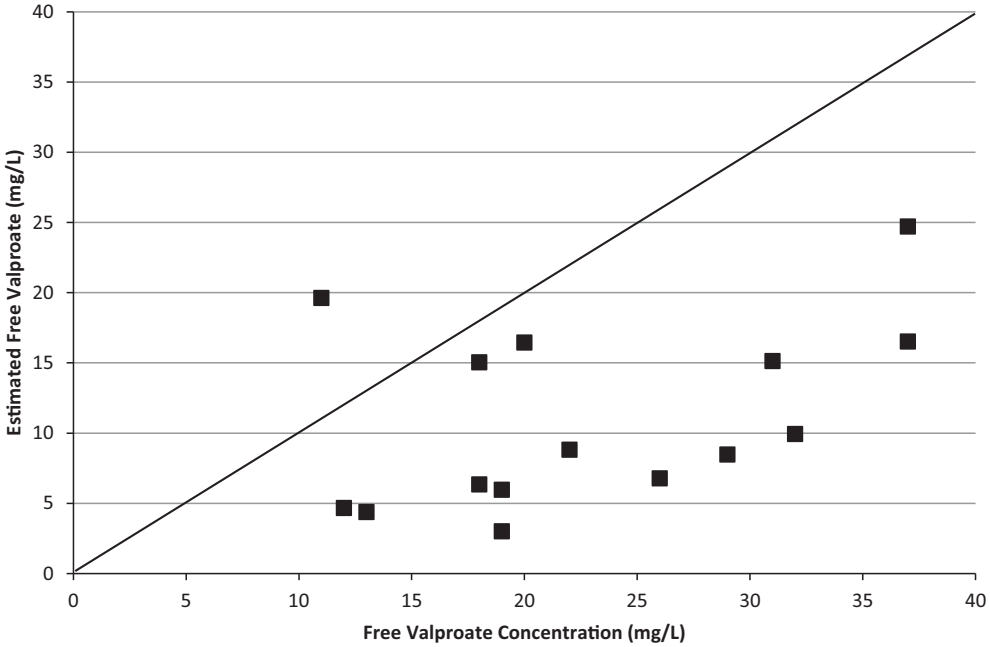

Figure 3. Free versus estimated-free serum valproate concentration calculated from albumin using Parent formula. Diagonal line = line of identity. The Parent formula underestimated free valproate concentrations (r = 0.45).

instead, the free fraction ranged from 15–89%. Monitoring total concentrations of medications is based on the assumption that they reflect free concentrations (i.e., that the free fraction is predictable and consistent between patients)[18]; this clearly was not the case for valproate among our cohort of ICU patients. Total serum valproate concentrations were usually low or in the reference range, yet the free concentration was usually above the reference range. The free fraction

was extremely variable, making any estimate based on the total concentration highly inaccurate. Even using a published formula to predict the free fraction and free concentration showed poor agreement with the actual measured values, perhaps because this formula was derived from non-ICU patients.[13] The complexity of valproate protein binding among critically ill patients suggests that monitoring actual free valproate concentrations may be necessary in ICU patients.

Our study represents the largest reported cohort of ICU patients with measured free serum valproate concentrations. Our data highlight the potential benefit of monitoring free serum valproate concentrations in critically ill patients, and increase the reported cases in the ICU literature nearly 5-fold. This finding is important because valproate is a preferred agent for seizure control and an emerging option for the management of ICU agitation.[8–10, 19] One report suggests that valproate may be more effective (75%) than phenytoin (59%) or levetiracetam (52%) to control status epilepticus.[20] With the recommendations to treat postcardiac arrest patients with seizures and myoclonus aggressively, it is likely that valproate use will increase in this patient population.[21]

Several clinical factors common in the ICU are associated with higher free fractions of valproate including uremia, hypoalbuminemia, free fatty acid administration (e.g., Intralipid, propofol, or clevidipine), and drug-drug interactions resulting in competition for albumin binding sites (e.g., aspirin and ibuprofen).[18, 22] Although documented more than 30 years ago, the potential utility of monitoring free serum valproate concentrations is not mentioned in clinical reviews or guidelines,[8–10, 23] and few ICU reports have been published.[4–7] More than 10 years ago, a formula to adjust total valproate concentrations was proposed because only 2% of laboratories were measuring free concentrations,[24] but our data suggest this formula is unreliable in ICU patients.

Older studies of outpatients treated with valproate concluded that monitoring free serum valproate concentrations was rarely helpful in patients without liver or renal disease or low albumin levels, showing free fractions in the expected range of 4–11%.[25, 26] They also showed no better relationship between seizure control or adverse effects with free versus total concentration monitoring for these patients.[25, 26] These data may have misled clinicians who work with critically ill patients at risk for altered protein binding. Elevated free serum valproate concentrations have been associated with ataxia, nystagmus, unsteady gait, and vomiting,[27] may be a stronger predictor of hyperammonemia than valproate dose or total concentration,[28] and may be inversely related to platelet count.[29]

Valproate exhibits concentration-dependent protein binding,[1] and in addition to the factors just described known to alter protein binding, the normal free fraction of 5–10% may increase to 30% or higher when total concentrations exceed 72 mg/L.[23] Only 1 of our 15 patients had a serum total concentration greater than 72 mg/L. As a low extraction ratio drug, valproate clearance depends on intrinsic hepatic clearance and free fraction; as free fraction increases, drug metabolism increases, potentially leading to lower than expected total serum concentrations, yet appropriate or even elevated free concentrations.[4, 11] Patients undergoing targeted temperature management may be especially at risk for unexpected pharmacokinetics because intrinsic hepatic clearance may be reduced.[30, 31]

Hypoalbuminemia is a common risk factor for an increased free fraction of valproate.[4, 6, 32, 33] The largest study published to date (including both hospitalized and outpatients) concluded that albumin values less than 3.5 g/L were associated with increased valproate free fractions, the only factor explaining therapeutic discordance between free and total serum valproate concentrations.[33] All patients in our study had albumin concentrations less than 3.7 g/dl. Uremia may also alter valproate protein binding, with free fractions of 20% with renal dysfunction compared with 8% without, and a moderate correlation has been observed between valproate free fraction and both serum creatinine and blood urea nitrogen.[34]

Free fatty acids can displace highly protein bound drugs due to their high binding affinity. Stearic, palmitic, oleic, and linoleic acid have been shown to increase the free fraction of valproate in a concentration-dependent fashion by 19–118%.[35] Propofol and Intralipid are rich in free fatty acids and have been shown to increase the free fraction of valproate.[36] Six of our patients were receiving propofol infusions. Although no patient in our study was receiving clevidipine, this dihydropyridine calcium channel blocker also contains free fatty acids and would likely increase the free fraction of valproate. Antipyretic doses of aspirin may result in a 4-fold increase in valproate free fraction, and similar effects have been seen with lower

salicylate doses (325 mg/day).[22, 37] Ibuprofen can also displace valproate, leading to decreasing total serum concentrations and the risk for inappropriate dose increases.[38, 39]

Several limitations of our case series warrant comment. These data are from a single hospital, but the types of patients reported and medications coadministered are not unique to our center. We collected data from a consecutive series of patients over 1 year, but unexpected biases are possible. Two different total valproate reference ranges are accepted: 50–100 mg/L for seizure management and 50–125 mg/L for behavioral issues. We selected the more inclusive 50–125 mg/L because some patients received valproate for behavioral issues. Although the range of 50–125 mg/L may have biased our sample toward more discordance, no patient had a total concentration higher than 100 mg/L.

The data supporting specific clinical effects and adverse events associated with discrete free serum valproate concentrations are not robust, which may be the greatest limitation of our report. Indeed, 80% of the five patients without adverse events had higher values for free and total valproate concentrations. Many different reference ranges for free serum valproate concentrations have been published (Table S1).[5–7, 26, 32, 40–42] The median and interquartile range from these reports identifies a low reference value of 5 mg/L (IQR 4.95–5 mg/L) and a high value of 12.9 mg/L (IQR 10–15.7 mg/L) similar to our reference range. Even if we used the highest reported range for free valproate concentrations as reported in the literature (6–20 mg/L), seven of our patients were above this threshold, and all were discordant (two low total concentrations and five within the reference range of 50–100 mg/L).

Conclusions

These cases highlight the great variability of valproate protein binding and free fraction among ICU patients and the potential problems associated with adjusting valproate doses based on total serum valproate concentrations. Estimating free valproate fraction or concentration by correcting for albumin level was similarly inaccurate in our ICU cohort. Monitoring free serum valproate concentrations may be appropriate for ICU patients with hypoalbuminemia, uremia, substances known to displace valproate from albumin (e.g., aspirin, ibuprofen, Intralipid, propofol, or clevidipine), total concentrations in the high end of the reference range (more than 72 mg/L), or unexpectedly low total serum valproate concentrations. Additional research to define best practice for dosing and monitoring valproate and to determine the relationship between free serum valproate concentrations and clinical and adverse effects in ICU patients is needed.

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Supporting Information

The following supporting information is available in the online version of this paper:

Figure S1. Altman-Bland plot showing Parent formula underestimates free serum valproate concentration, with bias of −11.9 mg/L and 95% limits of agreement of −28 to 4.2 mg/L.

Table S1. Literature Reported Reference Range for Free Valproate Concentrations