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Unbound Cephalothin Pharmacokinetics in Adult Burn Patients Are Related to the Elapsed Time after Injury▽

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Cephalothin (cefalotin) pharmacokinetics were evaluated for nine severely burned patients (42% ± 9% mean burn areas) and five healthy volunteers by using non-plasma-protein-bound concentration-time profiles. Burn patients gave increased mean residence times (36%) and reduced total clearances (25%). Mean residence times and distribution volumes increased between 1 and 4 days posttrauma, suggesting that burn patient pharmacokinetics change during the initial fluid resuscitation phase of treatment.

Interpatient variability is a consistent feature of burn patient pharmacokinetics (2). Fluid shifts between different body compartments, fluid resuscitation, and effects of surgery all have impacts on the pharmacokinetics of critically ill patients in a variable and unpredictable manner (10, 11). We recently reported that interstitial concentrations of cephalothin (cefalotin) were maintained significantly longer in burnt and nonburnt subcutaneous tissues of burn patients than in those of healthy individuals (4). In the current study, we examine the unbound plasma cephalothin pharmacokinetics of the same burnt and nonburnt individuals. The aim was to provide evidence in support of our published hypothesis that increased cephalothin exposure of burnt and nonburnt tissue sites is a consequence of severe burn injury and the management of shock by fluid resuscitation.

This study was an open-label, single-center pharmacokinetic study of cephalothin in patients with >28% body surface area (BSA) burns during burn debridement surgery within 4 days of trauma. Nine burn patients and five healthy volunteers were studied. The predicted theoretical benefit of prophylactic β-lactam use in this patient cohort has already been discussed (5), and interstitial concentrations of cephalothin have been reported previously for each volunteer and for six of the burn patients (4). The protocol received approval from the Hospital and University of Queensland Human Research Ethics Committees. Written informed consent was obtained from the legal guardians of the enrolled patients and from the healthy volunteers. Detailed descriptions of the research protocols have been published previously (4, 5). Briefly, patients received 1 g cephalothin in 10 ml 0.9% saline for 5 min through a dedicated lumen of a central venous catheter within 1 h of surgery commencement; volunteers received the same treatment via a forearm vein. Blood sampling into heparinized Vacutainers (BD, NJ) commenced upon the start of the infusion at stipulated time intervals (5, 10, 20, 30, 60, 120, and 240 min); plasma samples were stored at −20°C and analyzed within 1 week of collection. Methods for plasma analysis by isocratic high-performance liquid chromatography with UV detection and relevant assay validation details, have been published elsewhere (5). Unbound cephalothin data were calculated from total plasma cephalothin by correcting for percent plasma binding observed in prestudy plasma samples for each individual. Briefly, 500 μl of 100 μg/ml cephalothin in plasma was incubated (37°C for 30 min) and ultracentrifuged (12,000 × *g* for 20 min) through 30-kDa-nominal-cutoff membrane devices (Amicon YM30; Millopore Corporation, Billerica, MA) to give a filtrate yield of approximately 25% original volume, which was analyzed by high-performance liquid chromatography.

Unbound cephalothin plasma concentration-time data were analyzed using biexponential regression ($C = A\ e^{-\alpha t} + B\ e^{-\beta t}$), applying $1/y^2$ weighting to account for assay heteroscedasticity (1). Terminal elimination half-life ($t_{1/2\beta}$) was defined as 0.693/β, where β is the terminal elimination rate constant, taken from *C*. The maximal plasma cephalothin concentrations (C_{max}) were the observed values. Area under the curve (AUC) and area under the first moment curve (AUMC) for unbound cephalothin plasma profiles were calculated by the linear trapezoidal method and extrapolated to infinity, where $AUC_{0-\infty}$ is defined as $AUC_{0-t} + C_t/\beta$ and where $AUMC_{0-\infty}$ is defined as $AUMC_{0-t} + (t \cdot C_t/\beta) + C_t/\beta^2$. Total clearance ($CL_T$) was defined as dose/$AUC_{0-\infty}$ and was normalized for body weight. Mean residence time (MRT) was defined as $AUMC_{0-\infty}/AUC_{0-\infty}$ minus mean infusion time, which was defined as infusion time/2. Effective half-life ($t_{1/2}$) was defined as 0.693 · MRT. Apparent volume of distribution under steady-state conditions (V_{ss}) was defined as CL_T · MRT and was normalized for body weight. Statistical analysis was performed with SPSS version 16.0.1 for Windows (SPSS, Inc.), and the R computing language was used to compile plots (R Develop-

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TABLE 1. Patient demographics and health assessment scores

Patient	Sex	Age[a] (yr)	Ht[a] (m)	Wt[a,e] (kg)	BSA[a] (m^2)	TBSA (% 3rd/% 2nd)[c]	Score[a,b]		No. of days posttrauma	Concn (g/liter)[d]		
							APACHE II	SOFA		Plasma protein	Albumin	Globulin
1	Male	54	1.75	88	2.09	15/15	15	7	2	46	30	16
2	Male	25	1.80	70	1.88	26/27	14	5	2	44	27	17
3	Male	23	1.79	80	2.00	25/10	15	3	3	34	18	16
4	Male	42	1.80	80	2.01	38/10	15	5	4	44	22	22
5	Male	23	1.84	90	2.15	25/20	15	5	1	46	30	16
6	Male	50	1.73	90	2.10	12/16	13	9	3	47	29	18
7	Male	49	1.70	70	1.83	0/45	9	6	1	44	28	16
8	Male	23	1.78	100	2.24	51/3	21	7	1	30	18	17
9	Female	31	1.60	55	1.58	30/10	8	6	2	37	26	11
Mean (SD)		36 (12)	1.75 (0.07)	80 (13)	1.99 (0.19)	25/17 (14/12)	14 (4)	6 (2)	2 (1)	41 (6)	25 (5)	17 (2)

[a] Observations made upon admission to the intensive care unit.
[b] SOFA, sepsis related organ failure assessment; APACHE II, acute physiology and chronic health evaluation.
[c] TBSA assessment is itemized by wound depth (3rd, full thickness; 2nd, partial thickness) based on data from burn surgery records.
[d] Observations made upon the day of study.
[e] Estimated values.

ment Core Team [http://www.r-project.org/]). Nonpaired *t* tests with correction for unequal variance (verified by Levene's *f* test) were applied, except for $t_{1/2\beta}$ data, which, being homoscedastic, were analyzed without correction.

Demographics for the nine patients are given in Table 1, consisting of data compiled from intensive care unit records, including plasma protein analysis, sepsis-related organ failure assessment (12), and acute physiology and chronic health evaluation (8) data. Total burn surface area (TBSA), subcategorized as partial thickness (second-degree) and full thickness (third-degree) burns, was assessed by the same burn surgeon throughout. Total body surface area (BSA) was calculated by the method of Gehan and George (7). Patient and volunteer creatinine clearance (CL_{CR}) levels were estimated by Pathology Queensland (Herston, Queensland) from prestudy plasma samples and an 8-hour urine sample collection (3). The demographics for the five healthy volunteers (mean ± standard deviation [SD]) were as follows: age, 35 ± 10 years; height, 1.73 ± 0.12 meters; weight, 68 ± 13 kg; BSA, 1.82 ± 0.23 m^2.

The pharmacokinetics data are presented in Table 2. Patients with burn injury had a 45% increase in observed C_{max} ($P < 0.05$; *t* test), a 25% reduction of cephalothin CL_T ($P < 0.05$; *t* test), and a 36% increase in MRT ($P < 0.05$; *t* test) in comparison to healthy volunteers. Effective plasma $t_{1/2}$ was also increased in patients with burns in comparison to levels for healthy volunteers ($P < 0.05$; *t* test), reflecting the increased MRT since $t_{1/2}$ is 0.693 · MRT. Terminal elimination $t_{1/2\beta}$ had a trend of being longer in patients than in healthy volunteers that was not statistically significant ($P = 0.08$; *t* test). The differences between patients and healthy volunteers in $AUC_{0-\infty}$ and V_{ss} were not statistically significant. Lower percentages of plasma protein binding of cephalothin were observed for patients with burns than for healthy individuals (59% ± 8% versus 71% ± 2%; $P < 0.001$), and higher mean CL_{CR} values were recorded for patients with burns than for healthy individuals (148 ± 75 ml/min versus 98 ± 19 ml/min; $P < 0.001$; *t* test).

The dependent pharmacokinetic variables of MRT are V_{ss} and CL_T (since MRT is V_{ss}/CL_T), and accordingly, there were significant associations between MRT and both V_{ss} and $1/CL_T$ ($r = 0.727$ [$P < 0.05$] and $r = 0.798$ [$P = 0.01$], respectively). Aberrant pharmacokinetics in burn patients have previously been shown to exhibit covariance with amount of time following burn trauma (9), and in this study, MRT correlated positively with number of days posttrauma ($r = 0.727$; $P < 0.05$) (Fig. 1). Interestingly, V_{ss} also correlated with number of days

TABLE 2. Pharmacokinetics of unbound cephalothin in patients with severe burns and in a healthy volunteer control group

Patient	C_{max} (mg/liter)	$AUC_{0-\infty}$ (mg/h/liter)	MRT (h)	$t_{1/2}$ (h)	V_{ss} (liters/kg)	CL_T (liters/h/kg)	$t_{1/2\beta}$ (h)	CL_{CR} (ml/min)	% Protein binding
1	30.5	13.36	0.95	0.66	0.81	0.85	1.25	163	65
2	32.9	7.00	0.39	0.27	0.79	2.04	0.81	202	63
3	24.6	12.76	0.73	0.51	0.72	0.98	0.86	169	46
4	35.9	15.72	1.07	0.74	0.85	0.80	1.17	81	62
5	32.1	6.92	0.30	0.21	0.48	1.61	0.76	170	72
6	21.8	10.05	0.58	0.40	0.64	1.11	0.91	64	65
7	65.9	14.63	0.45	0.31	0.44	0.98	0.95	125	58
8	23.6	7.00	0.35	0.24	0.50	1.43	0.66	307	51
9	80.9	21.01	0.53	0.37	0.46	0.87	0.82	52	52
Mean (SD) for:									
Patients	38.7 (19.4)	12.05 (4.52)	0.59 (0.25)	0.41 (0.18)	0.63 (0.16)	1.19 (0.40)	0.91 (0.18)	148 (75)	59 (8)
Volunteers ($n = 5$)	21.3 (5.5)	9.89 (2.83)	0.38 (0.03)	0.26 (0.02)	0.60 (0.09)	1.58 (0.15)	0.74 (0.12)	98 (19)	71 (2)

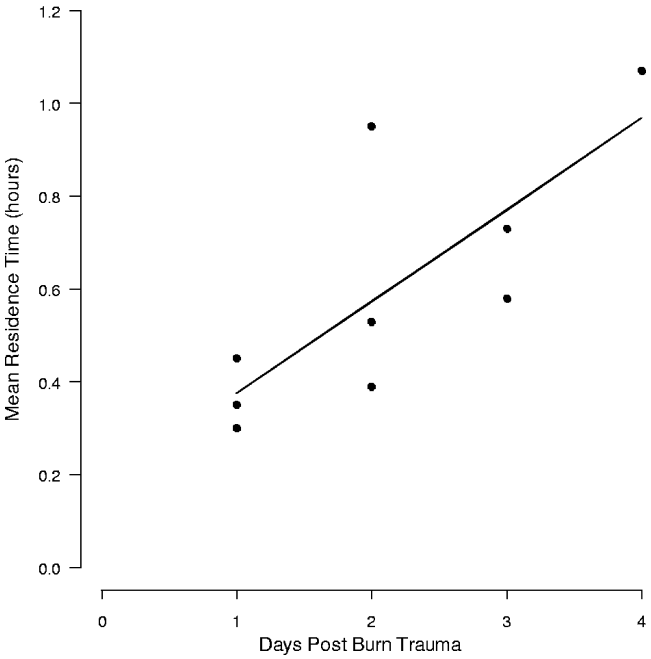

FIG. 1. Relationship between number of days following burn trauma and cephalothin (cefalotin) MRT for patients with severe burns. Linear fit: $y = 0.1975 \cdot x + 0.1775$.

posttrauma ($r = 0.724$; $P < 0.05$), but $1/CL_T$ did not ($r = 0.515$; nonsignificant), indicating that alterations in tissue distribution as opposed to changes in renal function underlie altered MRT in this burn patient cohort.

We previously reported increased exposure of both burnt and nonburnt subcutaneous tissues to cephalothin in burn patients compared with healthy volunteers (4). This increased peripheral-tissue residence time appears to be reflected in the MRT calculated from unbound plasma data. The cumulative effects on tissue fluid volume of systemic inflammatory response syndrome, net fluid loading, and impaired lymphatic drainage build up during the resuscitative phase of burn treatment. We have shown that MRT and its dependent variable V_{ss} also increase during this period. We conclude that tissue pharmacokinetics play a decisive role in determining unbound plasma pharmacokinetics in burn patients and speculate that aberrant antimicrobial pharmacokinetics may be an unanticipated consequence of excessive fluid resuscitation (6) or an effect of the inflammatory response itself (11).

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REFERENCES

1. **Almeida, A. M., M. M. Castel-Branco, and A. C. Falcão.** 2002. Linear regression for calibration lines revisited: weighting schemes for bioanalytical methods. J. Chromatogr. B **774:**215–222.
2. **Blanchet, B., V. Jullien, C. Vinsonneau, and M. Tod.** 2008. Influence of burns on pharmacokinetics and pharmacodynamics of drugs used in the care of burn patients. Clin. Pharmacokinet. **47:**635–654.
3. **Cockcroft, D. W., and M. H. Gault.** 1976. Prediction of creatinine clearance from serum creatinine. Nephron **16:**31–41.
4. **Dalley, A. J., J. Lipman, R. Deans, B. Venkatesh, M. Rudd, M. S. Roberts, and S. E. Cross.** 2009. Tissue accumulation of cephalothin in burns: a comparative study by microdialysis of subcutaneous interstitial fluid cephalothin concentrations in burn patients and healthy volunteers. Antimicrob. Agents Chemother. **53:**210–215.
5. **Dalley, A. J., J. Lipman, B. Venkatesh, M. Rudd, M. S. Roberts, and S. E. Cross.** 2007. Inadequate antimicrobial prophylaxis during surgery: a study of beta-lactam levels during burn debridement. J. Antimicrob. Chemother. **60:**166–169.
6. **Dulhunty, J. M., R. J. Boots, M. J. Rudd, M. J. Muller, and J. Lipman.** 2008. Increased fluid resuscitation can lead to adverse outcomes in major-burn injured patients, but low mortality is achievable. Burns **34:**1090–1097.
7. **Gehan, E. A., and S. L. George.** 1970. Estimation of human body surface area from height and weight. Cancer Chemother. Rep. **54:**225–235.
8. **Knaus, W. A., E. A. Draper, D. P. Wagner, and J. E. Zimmerman.** 1985. APACHE-II: a severity of disease classification-system. Crit. Care Med. **13:**818–829.
9. **Mohr, J. F., L. Ostrosky-Zeichner, D. J. Wainright, D. H. Parks, T. C. Hollenbeck, and C. D. Ericsson.** 2008. Pharmacokinetic evaluation of single-dose intravenous daptomycin in patients with thermal burn injury. Antimicrob. Agents Chemother. **52:**1891–1893.
10. **Pinder, M., R. Bellomo, and J. Lipman.** 2002. Pharmacological principles of antibiotic prescription in the critically ill. Anaesth. Intensive Care **30:**134–144.
11. **Roberts, J. A., and J. Lipman.** 2006. Antibacterial dosing in intensive care—pharmacokinetics, degree of disease and pharmacodynamics of sepsis. Clin. Pharmacokinet. **45:**755–773.
12. **Vincent, J. L., R. Moreno, J. Takala, S. Willatts, A. DeMendonca, H. Bruining, C. K. Reinhart, P. M. Suter, and L. G. Thijs.** 1996. The SOFA (sepsis-related organ failure assessment) score to describe organ dysfunction/failure. Intensive Care Med. **22:**707–710.