Anesthesiology 2002; 97:1082–92

Concentration–Effect Relation of Succinylcholine Chloride during Propofol Anesthesia

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Background: **The pharmacokinetics and pharmacodynamics of succinylcholine were studied simultaneously in anesthetized patients to understand why the drug has a rapid onset and short duration of action. A quantitative model describing the concentration–effect relation of succinylcholine was proposed. The correlation between *in vitro* hydrolysis in plasma and *in vivo* elimination was also examined.**

Methods: **Before induction of anesthesia, blood was drawn for *in vitro* analysis in seven adults. Anesthesia was induced with propofol and remifentanil. Single twitch stimulation was applied at the ulnar nerve every 10 s, and the force of contraction of the adductor pollicis was measured. Arterial blood was drawn frequently after succinylcholine injection to characterize the front-end kinetics. Plasma concentrations were measured by mass spectrometry, and pharmacokinetic parameters were derived using compartmental and noncompartmental approaches. Pharmacokinetic–pharmacodynamic relations were estimated.**

Results: **The mean *in vitro* degradation rate constant in plasma ($1.07 \pm 0.49\ min^{-1}$) was not different from the *in vivo* elimination rate constant ($0.97 \pm 0.30\ min^{-1}$), and an excellent correlation ($r^2 = 0.94$) was observed. Total body clearance derived using noncompartmental ($37 \pm 7\ ml \cdot min^{-1} \cdot kg^{-1}$) and compartmental ($37 \pm 9\ ml \cdot min^{-1} \cdot kg^{-1}$) approaches were similar. The plasma–effect compartment equilibration rate constant (k_{eo}) was $0.058 \pm 0.026\ min^{-1}$, and the effect compartment concentration at 50% block was 734 ± 211 ng/ml.**

Conclusion: **Succinylcholine is a low-potency drug with a very fast clearance that equilibrates relatively slowly with the effect compartment. Its *in vivo* disappearance is greatly accountable by a rapid hydrolysis in plasma.**

SUCCINYLCHOLINE chloride, a depolarizing neuromuscular blocking agent, has been extensively used in anesthesia since 1951. In most patients with no qualitative or quantitative plasma cholinesterase deficiency, succinylcholine has the advantage of a rapid onset and short duration of action, two characteristics that are extremely valuable, especially in the full-stomach, emergency situation. It is generally recognized that the short time course of succinylcholine blockade is due to its rapid metabolism, but other factors could play a role. The search for the ideal nondepolarizing drug involves an understanding of the factors that makes succinylcholine blockade so evanescent. This entails simultaneous measurement of plasma concentrations (pharmacokinetics) and neuromuscular blockade (pharmacodynamics).

Succinylcholine is known to be hydrolyzed by plasma cholinesterase.[1] Genetic variants of the plasma cholinesterases, low plasma cholinesterase activity due to an acquired deficiency (liver disease, carcinoma, debilitating disease, and so forth), and other factors, among which some remain unknown, may lead to an episode of prolonged apnea after succinylcholine.[2] Because of this recognized variation in the activity of plasma cholinesterases, large interpatient differences in pharmacokinetic, pharmacodynamic, and pharmacokinetic–pharmacodynamic relations are expected.

For drugs with a fast onset of effect, the importance of obtaining frequent samples during the first minutes after a bolus injection has been demonstrated.[3,4] In addition, a pharmacokinetic–pharmacodynamic analysis must be performed by measuring the drug effect simultaneously. Pharmacokinetic studies of succinylcholine have been hindered by the lack of a suitable assay due to the analytical challenge involved with measuring its concentration in plasma. Only one human pharmacokinetic study has been reported,[5] and it did not include pharmacokinetic–pharmacodynamic relations. Recently, an electrospray tandem mass spectrometry method that is 80 times as sensitive was developed in our laboratory.[6]

The primary objective of this study was to provide a quantitative model to describe the kinetics and the dynamics of succinylcholine chloride after a 1-mg/kg bolus dose in anesthetized patients. The secondary objective was to determine if the *in vitro* rate of degradation in plasma is of predictive value for the *in vivo* elimination rate of succinylcholine in a given patient.

Materials and Methods

Patients

The study protocol was approved by the Research Ethics Committee at the Centre Hospitalier de l'Université de Montréal. Written informed consent was obtained from each subject. Patients included were adults, aged 18–65 yr, with American Society of Anesthesiologists status I or II who were scheduled for surgery for which an arterial cannula was indicated. Patients with cardiovascular, pulmonary, neuromuscular, hepatic, or renal disease were excluded. Other exclusion criteria were the concurrent

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Received from the Faculté de Pharmacie, Université de Montréal, Département d' Anesthésiologie, Centre Hospitalier de l' Université de Montréal, Montréal, Québec, Canada. Submitted for publication October 25, 2001. Accepted for publication May 30, 2002. Supported by grants No. MA-10274 (to Drs. Donati and Varin) and MT-6712 (to Dr. Mamer) from the Canadian Institutes of Health Research, Ottawa, Ontario, Canada. Presented in part at the annual meeting of the American Society of Anesthesiologists, New Orleans, Louisiana, October 14–17, 2001.

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administration of drugs known to be or suspected of interfering with the neuromuscular function, anemia, and body mass index less than 19 or greater than 29.9. Patients with a personal or familial history of an abnormal response to succinylcholine or mivacurium were excluded.

Once patients arrived in the operating room, a cannula was inserted into the left radial artery before induction of anesthesia. Monitoring included continuous electrocardiography, pulse oximetry, and an automatic noninvasive blood pressure device. Anesthesia was induced with remifentanil (0.5–1 μg/kg) and propofol (2–3 mg/kg) to provide sufficient depth of anesthesia for tracheal intubation without neuromuscular blocking agents. Mechanical ventilation was adjusted to keep end-tidal carbon dioxide in the range of 30–35 mmHg. Anesthesia was maintained with 100–200 $\mu g \cdot kg^{-1} \cdot min^{-1}$ propofol, 0.1–0.2 $\mu g \cdot kg^{-1} \cdot min^{-1}$ remifentanil, and oxygen. After stabilization of vital signs and neuromuscular function, a 1-mg/kg bolus dose of succinylcholine chloride was administered over 2 s. If a patient required maintenance of neuromuscular block after the first dose of succinylcholine, vecuronium was given after at least 75% of twitch recovery relative to presuccinylcholine control was reached. The study was terminated when the last plasma sample was collected and at least 75% of neuromuscular recovery from the baseline value was reached.

Neuromuscular Monitoring

After induction of anesthesia, the mechanomyographic response of the adductor pollicis muscle to single twitch (0.2 ms duration and frequency of 0.1 Hz) of the ulnar nerve at the right wrist was measured with a Grass FT10 force displacement transducer (Grass Instruments, Quincy, MA). A stabilization period of at least 3 min was allowed before injection of succinylcholine (reference value for onset). Muscle relaxation was monitored continuously until neuromuscular tension had returned to more than 75% of its baseline value. All patients but one (patient no. 1) recovered completely (reference value for recovery) before the administration of another neuromuscular blocking agent.

Blood Sampling Schedule

Before induction of anesthesia, an arterial blood sample (48 ml) was drawn from each patient into heparinized tubes to determine the patients' phenotype and the *in vitro* rate of degradation of succinylcholine in plasma. Another sample was drawn (1.5 ml) into EDTA and echothiophate (20 μl of a 40-mM solution per milliliter of plasma) enriched Vacutainer tube for the *in vivo* pharmacokinetic profile (blank sample). After the 1-mg/kg intravenous bolus dose of succinylcholine chloride, arterial blood samples (1.5 or 3 ml) were drawn into tubes containing heparin (20 U) and echothiophate. Echothiophate was added to prevent the hydrolysis of succinylcholine by plasma cholinesterase.[7] For the first patient, arterial blood samples were drawn every 5 s for the first 2 min (1.5 ml each) and at 3, 4, 5, 7, 10, 15, 20, and 25 min (3 ml each). For this patient, succinylcholine was not detectable after 7 min. Therefore, for the other patients, the sampling schedule was the same for the first 2 min, then blood samples were drawn every minute until 10 min (3 ml each). During the first 2 min, sampling was performed by letting blood come out under arterial pressure and switching collecting tubes every 5 s. The midpoint of collection was used as the recorded time point for the pharmacokinetic analysis. To minimize the *ex vivo* degradation of succinylcholine, blood samples were kept in an ice-water bath and centrifuged within 5 min. The *in vivo* plasma samples were frozen on dry ice immediately thereafter and were stored at −70°C until analysis. The plasma samples obtained for *in vitro* analysis was kept in an ice-water bath until processing.

Phenotyping of Patients' Plasma

Each patient's phenotype was characterized using the conventional laboratory methods, by measurement of cholinesterase activity and biochemical inhibition reactions, such as dibucaine number, fluoride number, and chloride number.[8] These analyses were conducted in the biochemistry laboratory at Maisonneuve-Rosemont Hospital.

In Vitro *Degradation Study*

The *in vitro* study was performed within 4 h of blood collection to ensure the integrity of the medium. Incubations in fresh plasma were conducted at the patient's body temperature as measured in the operating room (36 ± 1°C). Succinylcholine was added to the patient's plasma to achieve a final concentration of 2 μg/ml and, immediately after, 1 ml of the incubation mixture was removed and stabilized with echothiophate. This was designated as time zero for the incubation. Subsequent plasma samples were treated similarly at 2, 3, 4, 5, 7, 10, 15, 20, and 25 min for the first patient and at 0.5, 1, 1.5, 2, 3, 4, 5, 6, 7, 8, 9, and 10 min for all other patients. Samples were flash frozen on dry ice and stored at −70°C until analysis.

Analysis of Samples

Succinylcholine was extracted from human plasma on C1 solid-phase cartridges and was analyzed using positive ion electrospray tandem mass spectrometry (ESI/MS/MS) after direct probe injection.[6] Briefly, plasma concentrations were determined with a stable isotope dilution assay using hexadeuterosuccinylcholine as the internal standard. The calibration curve was prepared using the ratio of intensities of the major product ions; m/z = 115.5 for succinylcholine and m/z = 117.0 for the internal standard. Calibration curves were linear

from 25 to 4,000 ng/ml. Dilution of patient samples for concentrations exceeding 4,000 ng/ml was validated using dilution of quality control samples. For intraday precision, coefficient of variation was 6% or less, and accuracy ranged from 98 to 103%. For the interday precision, coefficient of variation was 10% or less, and accuracy ranged from 90 to 102%. The method was specific as each patient's blank plasma sample was free of interference in the spectral area of interest.[6]

Pharmacokinetic Analysis

In the design of our study, we had hypothesized that the *in vitro* speed of degradation could be related to the *in vivo* elimination half-life of succinylcholine. As the *in vitro* rate constant is usually determined with a noncompartmental model, a similar approach was also conducted for the *in vivo* study. The results of the noncompartmental model served also to validate the results obtained with the compartmental approach. All pharmacokinetic analyses were performed using raw data only. Plasma concentration *versus* time profiles were analyzed using WINNONLIN (Professional Edition version 1.5; Pharsight, Mountain View, CA). Point estimates and pharmacokinetic parameters were optimized using a standard minimization method (a Levenberg and Hartley modification of the Gauss-Newton algorithm).[9,10]

Noncompartmental Approach

The area under the plasma concentration–time curve after intravenous administration of succinylcholine was calculated by the linear trapezoidal rule method. Lag time was defined as the time between injection of succinylcholine and the time when the last null concentration was observed. The *in vitro* and the *in vivo* rate constants ($k_{in\ vitro}$ and k_{el}, respectively) were obtained from the least-square fitted log-linear terminal portion of the plasma concentration–time profile. The mean number of points, in the terminal portion of the curve, used for the determination of $k_{in\ vitro}$ was 6 (range, 5–7 points) and for k_{el} was 7 (range, 4–14 points). Total body clearance and the volume of distribution at steady state (V_{ss}) were calculated using standard formulae.[11]

Compartmental Approach

Pharmacokinetic parameters were also derived using a two-compartment model with central elimination only.[12,13] Traditional models assume an instantaneous input function after bolus administration. To account for the delay before mixing in the central compartment and to adequately characterize the intravascular mixing phase, the compartmental model included a lag time and a first-order input function (k_{01}), similar to an absorption phase. According to the Akaike information criterion, the pharmacokinetics of succinylcholine was better described by a two-compartmental model than by a one-compartment model.[14] A weighting function of 1/(predicted y^2) was applied. The following parameters were determined: the input rate constant to the central compartment (k_{01}), the distribution (α) and elimination (β) rate constants and their corresponding A and B coefficients, the volume of the central compartment (V_1), the intercompartmental clearances (Cl_{12} and Cl_{21}), the total clearance, the transfer rate constant from the central to peripheral compartment (k_{12}), the transfer rate constant from peripheral to central compartment (k_{21}), the exit rate constant from central compartment (k_{10}), and the volume of distribution at steady state assuming central elimination only ($V_{ss,c}$).

Succinylcholine undergoes the same metabolic pathway as mivacurium, and as much as 40% of the latter was recently found to be hydrolyzed during its passage through the forearm in humans.[15] Therefore, we could not exclude the possibility of an elimination from both the central and peripheral compartment (k_{20}) for succinylcholine as this, most importantly, may lead to an underestimation of the V_{ss}. Therefore, the V_{ss} when assuming both central and peripheral elimination ($V_{ss,c+p}$) was also calculated. In the absence of measured peripheral concentrations, k_{20} cannot be derived independently without making some assumptions.[16] We have previously proposed two approaches to account for peripheral elimination: K_{20} can either be assumed to be equal to $k_{in\ vitro}$ obtained from each patient's plasma or equal to β if it has already been shown that $k_{in\ vitro}$

Table 1. Demographic Data

Patient No.	Sex	Age (yr)	Weight (kg)	ASA Status	Surgical Procedure	Cholinesterase Activity* (U/l)	Inhibition Index Using			Phenotype
							Dibucaine No.† (U/l)	Fluoride No.‡ (U/l)	Chloride No.§ (U/l)	
1	M	52	80	II	Colectomy	20.2	0.71	0.59	0.14	Normal
2	M	56	71	I	Radical prostatectomy	28.8	0.54	0.49	0.15	Intermediate
3	F	36	56	I	Nephrectomy	NA	NA	NA	NA	NA
4	M	42	80	II	Femoro-popliteal graft	68.7	0.78	0.65	0.16	Normal
5	M	50	88	I	Hip prosthesis	57.6	0.75	0.66	0.08	Normal
6	F	48	77	II	Nephrectomy	39.9	0.78	0.61	0.12	Normal
7	M	49	68	I	Nephrectomy	28.8	0.73	0.56	0.18	Normal

* Normal range, 43.0–69.0 U/l; † Normal range, 0.78–0.85 U/l; ‡ Normal range, 0.57–0.64 U/l; § Normal range, 0.11–0.20 U/l.

ASA = American Society of Anesthesiologists.

Table 2. Pharmacodynamic Parameters

Patient No.	Onset (min)	Time to 25% Recovery (min)	Time to 50% Recovery (min)	Time to 75% Recovery (min)	Recovery Index 25–75% (min)
1	1.77	15.1	16.8	18.7	3.6
2	1.43	12.6	13.6	14.5	1.9
3	1.22	6.9	8.0	9.3	2.5
4	1.62	7.5	8.4	9.2	1.7
5	1.40	7.3	8.2	9.3	2.0
6	1.00	6.8	7.8	8.8	2.0
7	0.80	7.8	8.7	9.7	1.9
Mean	1.32	9.1	10.2	11.3	2.2
SD	0.34	3.3	3.6	3.8	0.6

equals β in a given patient.[13] The value that would be substituted for K_{20} would be decided according to the results obtained *in vivo* and *in vitro*.

Pharmacokinetic–Pharmacodynamic Analysis

The pharmacokinetic–pharmacodynamic analysis were conducted using two approaches: a nonparametric and a two-stage parametric approach. For both approaches, the pharmacodynamic parameters were determined using each patient's data separately.

For the nonparametric approach, the effect compartment equilibration rate constants, k_{e0}, was determined using the nonparametric method of Unadkat *et al.*[17] This approach makes no assumptions about the pharmacodynamic or the pharmacokinetic model while establishing a link between the plasma concentration and effect data. The optimal k_{e0} was found by successive iterations and defined as the value that minimized the average of squared distances between the ascending and descending limbs of the hysteresis loop. During the first 2 min, collection of blood samples was more frequent than twitch data (every 5 *vs.* 10 s). As the nonparametric approach requires the input of both plasma concentration and neuromuscular twitch at a given time, linear interpolation had to be used to generate twitch results during onset. Also, because the effect persisted while the plasma concentrations were below the limit of quantification, corresponding plasma concentrations were extrapolated using the terminal slope. A sigmoid E_{max} model was used to correlate the effect, with the effect-compartment concentration, thus providing values for the effect- compartment concentration at 50% blockade (EC_{50}) and the slope factor (γ).[17–19] The goodness of fit was assessed by the Akaike information criterion.

For the parametric approach, a sequential method was used. The pharmacokinetic parameters obtained with the compartmental analysis were fixed for the pharmacokinetic–pharmacodynamic modeling. A parametric link model was used to derive the equilibrium rate constant (k_{e0}) between the central compartment and the effect compartment using WINNONLIN (Professional Edition version 1.5; Pharsight). The EC_{50} and γ were determined using the sigmoid E_{max} model.[20] A weighting function of 1 was applied. Goodness of fit was assessed by the Akaike information criterion.

Statistical Analysis

During the model discrimination process, the predicted data were compared with the observed data by visual inspection of predicted concentration–time pro-

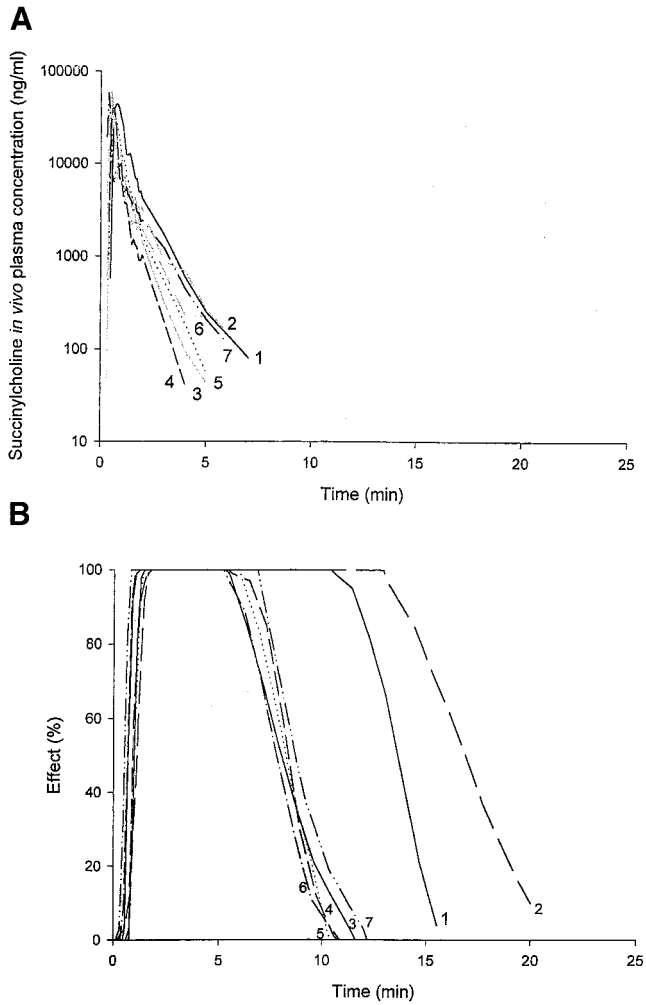

Fig. 1. (*A*) Individual succinylcholine *in vivo* plasma concentration-*versus*-time curves for the seven patients who received 1 mg/kg succinylcholine chloride. (*B*) Individual neuromuscular effect of succinylcholine for the seven patients.

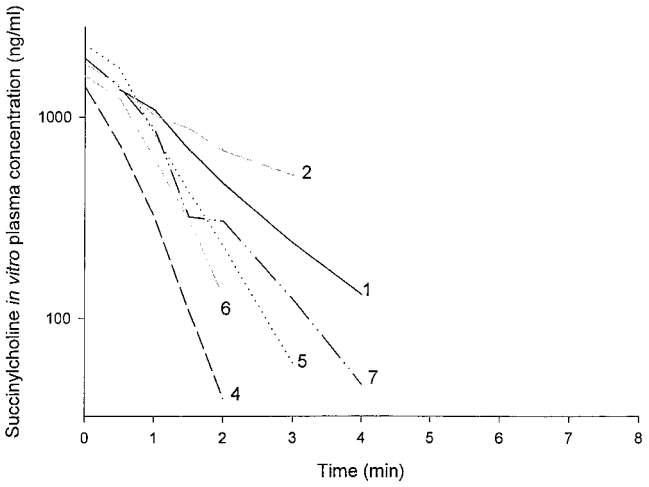

Fig. 2. Individual succinylcholine *in vitro* speed of degradation in plasma for six patients.

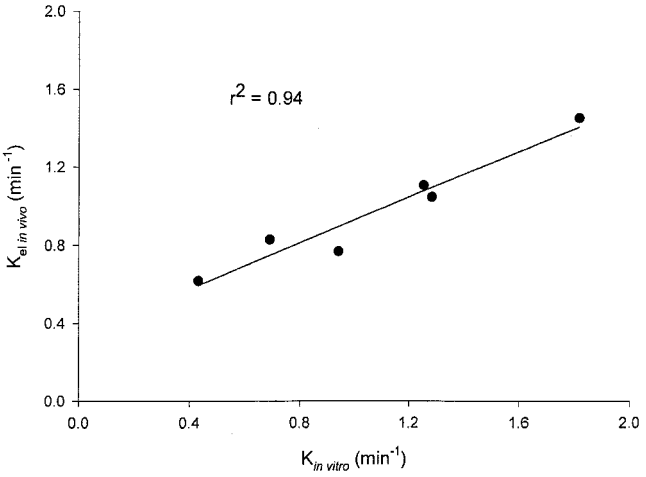

Fig. 3. Linear relation between the *in vitro* speed of degradation ($k_{in\ vitro}$) and the *in vivo* elimination constant (k_{el}) for six patients.

file, of weighted residuals, and by comparing the values of the Akaike information criterion obtained with either model.

Data are presented as mean values ± SD. For each patient, a paired *t* test or a Wilcoxon signed rank test when normality test failed was used to compare pharmacokinetic and pharmacokinetic–pharmacodynamic parameter estimates obtained with different models. The threshold for statistical significance (α) was set at 0.05.

Results

Demographic data are presented in table 1. A mean age of 48 ± 7 yr and mean weight of 74 ± 10 kg were observed for the seven patients recruited. In all but one patient, the phenotype was considered normal (table 1). Results for patient no. 3 are not available because the *in vitro* blood sample was lost. This precluded from obtaining the patient's phenotype results and also the *in vitro* degradation half-life.

Pharmacodynamic parameters are presented in table 2. All patients reached 100% neuromuscular block, and the onset time ranged from 0.8 to 1.8 min. Excluding 0% and 100% blockade, we observed 3–5 pharmacodynamic data points during the onset phase. The mean 25–75% recovery index ranged from 1.7 to 3.6 min.

The *in vivo* concentration–time profiles of succinylcholine in each patient, as well as the effect as a function of time, are presented in figure 1. The first detectable concentration was observed between 17.5 and 27.5 s. Peak concentrations were reached between 22.5 and 47.5 s and ranged from 30,378 to 56,906 ng/ml. Then, a rapid decrease in concentrations was observed. Recovery occurred after plasma concentrations of succinylcholine were no longer detectable.

In figure 2, the plasma concentration–time profiles for the *in vitro* incubation of succinylcholine are presented for six of seven patients. Using pairwise comparisons, the *in vitro* degradation rate constant ($k_{in\ vitro}$) and the *in vivo* elimination rate constant (k_{el}) obtained for each patient were not significantly different. The mean ± SD values for $k_{in\ vitro}$ and k_{el} were 1.07 ± 0.49 min^{-1} (corresponding to a half-life of 39 s) and 0.97 ± 0.30 min^{-1} (corresponding to a half-life of 43 s), respectively. When corresponding k_{el} and $k_{in\ vitro}$ values were plotted against each other, an excellent linear correlation (r^2 = 0.94) was observed (fig. 3).

Table 3. Compartmental Pharmacokinetic Results

						Descriptive		
Patient No.	C_{max} (ng/ml)	T_{max} (min)	Lag (min)	A (ng/ml)	B (ng/ml)	Alpha (min^{-1})	Beta (min^{-1})	k_{01} (min^{-1})
1	45,733	0.70	0.53	90,185	8,852	3.02	0.74	10.71
2	31,137	0.38	0.15	201,726	10,231	3.94	0.67	5.55
3	55,143	0.48	0.38	550,860	9,633	9.11	1.21	11.64
4	44,769	0.54	0.44	171,814	7,536	7.50	1.45	14.29
5	28,635	0.62	0.43	194,028	9,589	4.69	1.12	6.54
6	70,863	0.43	0.36	169,564	10,325	8.66	1.05	25.00
7	90,621	0.33	0.29	718,790	10,578	22.45	0.82	30.84
Mean	52,414	0.50	0.37	299,567	9,535	8.48	1.01	14.94
SD	22,094	0.13	0.12	236,879	1,054	6.60	0.28	9.50

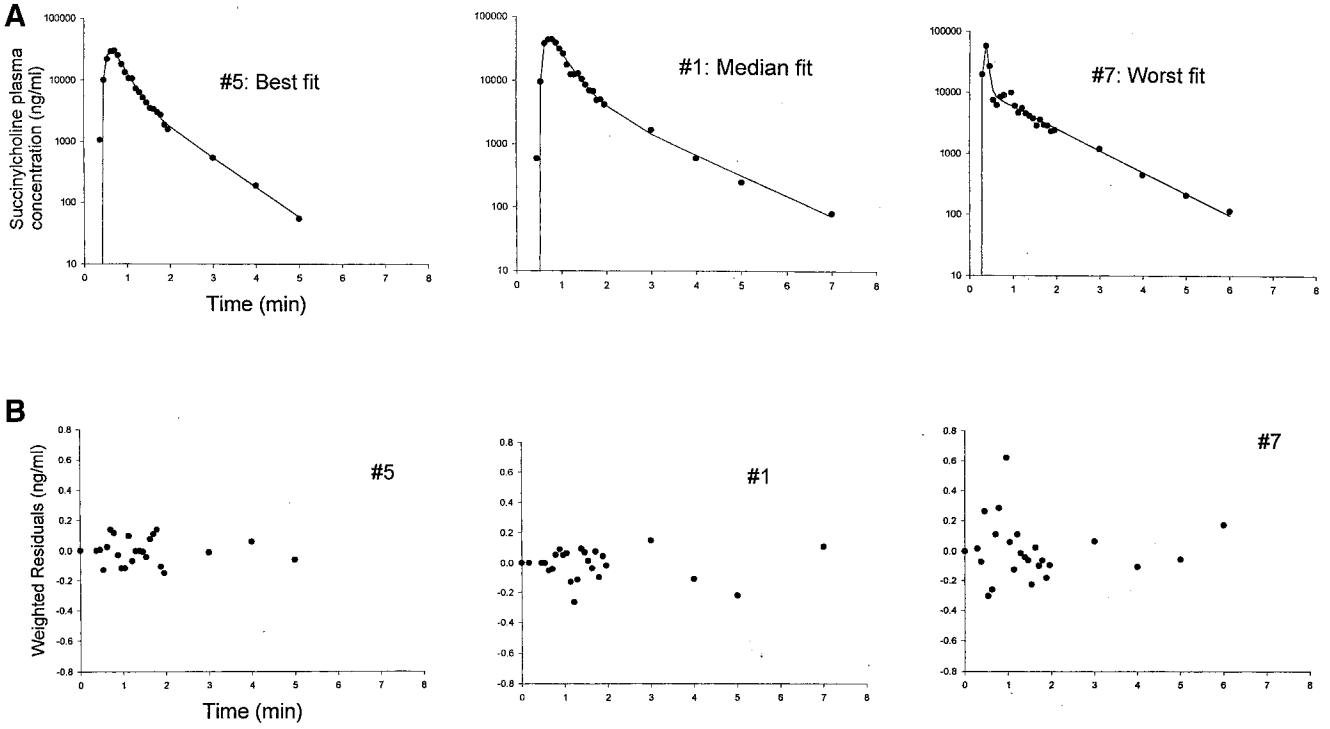

Fig. 4. (*A*) Observed *versus* predicted plasma concentration for patients no. 1, 5, and 7 using compartmental approach (central elimination only). (*B*) Their respective plots of weighted residuals.

The results from the compartmental pharmacokinetic analysis are presented in table 3. First, there was no significant difference between the maximum concentration (C_{max}), time when C_{max} occurred (T_{max}), and lag time observed and those estimated by the compartmental analysis. The mean observed C_{max}, T_{max}, and lag time were 49,182 ± 11,236 ng/ml, 0.57 ± 0.15 min, and 0.26 ± 0.07 min, respectively. A distribution phase with a mean half-life of 0.08 min (5 s) followed by a fast elimination phase with a mean half-life of 0.69 min (41 s) was observed. Mean total body clearance was similar for the compartmental (37 ± 9 ml · min^{-1} · kg^{-1}; table 3) and the noncompartmental (37 ± 7 ml · min^{-1} · kg^{-1}) approaches. Likewise, the mean elimination rate constants (k_{el} for the noncompartmental approach and β for the compartmental approach) were not different from each other, the mean noncompartmental elimination rate constant being 1.01 ± 0.29 min^{-1}. The V_{ss} calculated with the noncompartmental approach (20 ± 7 ml/kg) also gave a similar value to that obtained with the compartmental approach ($V_{ss,c}$). However, a twofold increase in V_{ss} was observed for the compartmental approach assuming both central and peripheral elimination ($V_{ss,c+p}$). As no statistical difference was observed between $k_{in\ vitro}$ and β for succinylcholine, $V_{ss,c+p}$ was calculated assuming that $k_{20} = \beta$.

Figure 4 shows the *in vivo* plasma concentration–time curves in three patients having the best, worst, and median fit as well as the respective plots showing the distribution of the weighted residuals. Likewise, the observed and predicted effects for these three patients are shown in figure 5.

An anticlockwise hysteresis was observed in the plasma concentration–effect relation of succinylcholine,

Table 3. (*continued*)

Parameters								
k_{10} (min^{-1})	k_{12} (min^{-1})	k_{21} (min^{-1})	Cl_{12} (ml · min^{-1} · kg^{-1})	Cl_{21} (ml · min^{-1} · kg^{-1})	Cl_t (ml · min^{-1} · kg^{-1})	V_1 (ml/kg)	$V_{ss,\ C}$ (ml/kg)	$V_{ss,\ C\ +\ P}$ (ml/kg)
2.2	0.5	1.0	6	6	25	11	17	34
2.4	1.1	1.1	13	13	28	12	24	45
6.3	2.3	1.7	14	14	40	6	14	34
5.7	1.3	1.9	12	12	51	9	15	36
3.3	0.9	1.6	12	12	43	13	20	41
5.4	2.6	1.7	17	17	36	7	17	35
9.7	11.7	1.9	46	46	38	4	28	48
5.0	2.9	1.6	17	17	37	9	19	39
2.6	3.9	0.4	13	13	9	3	5	6

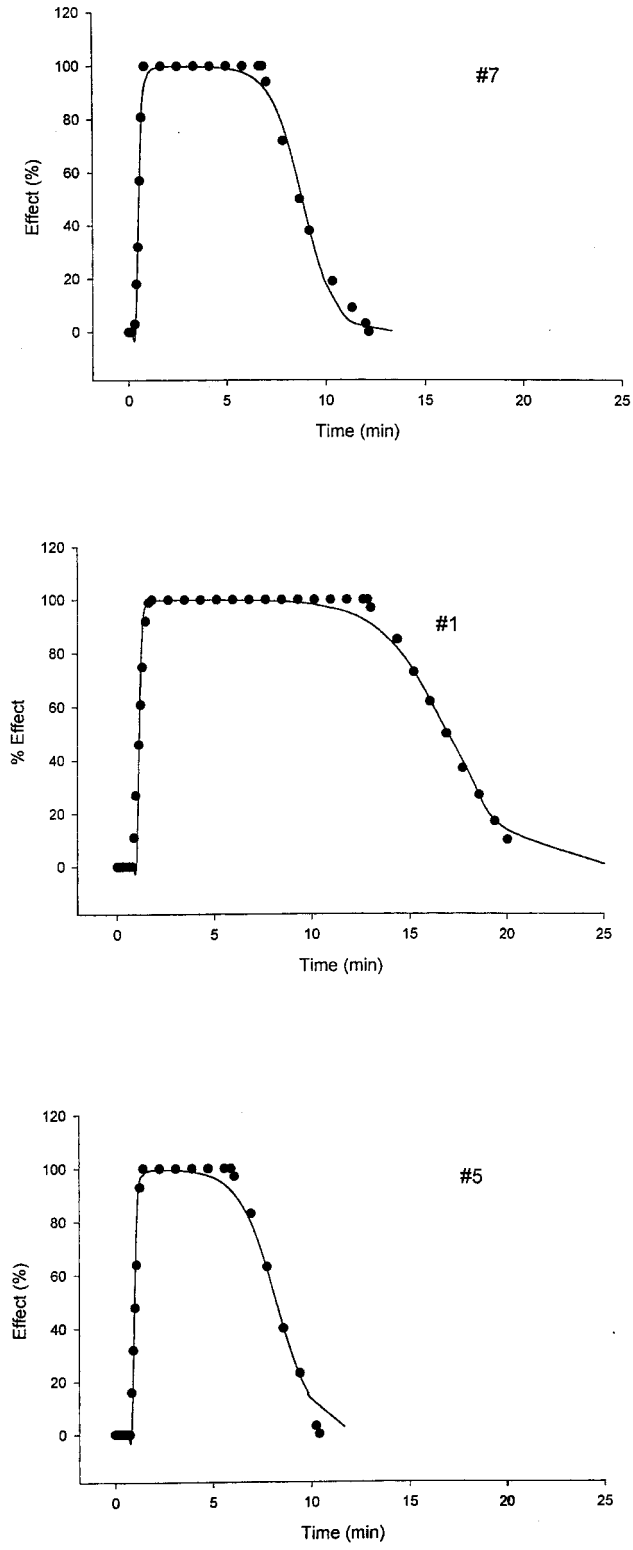

Fig. 5. Observed *versus* predicted neuromuscular effect for patients no. 1, 5, and 7 using the compartmental approach.

and a link model had to be used to simulate effect compartment concentrations before a sigmoid E_{max} model could be applied. Both the k_{e0} and EC_{50} values were not significantly different when derived by either the nonparametric or parametric approach (table 4). However, γ was twice as large with the parametric approach. The standard deviations of the EC_{50}, k_{e0}, and γ parameter estimates for individual patients ranged between 1 and 11%. When the onset times were plotted against their respective k_{e0} values, a strong linear correlation ($r^2 = 0.9$) was found (fig. 6).

Discussion

This study confirms the fast elimination half-life of succinylcholine and shows that, for each patient, the *in vivo* elimination half-life correlates strongly with the *in vitro* speed of degradation. The pharmacokinetic–pharmacodynamic analysis reveals that succinylcholine has a k_{e0} value comparable with that of most nondepolarizing agents and an EC_{50} denoting a low potency. Thus, our results suggest that the rapid onset and offset of succinylcholine neuromuscular block are due to a short plasma half-life, not to a fast access to muscle.

To our knowledge, only one pharmacokinetic study has been reported for succinylcholine in humans.[5] In that study, a bolus dose of 1 or 2 mg/kg of succinylcholine chloride was used. The mean values for clearance for both doses (700 and 250 ml · min^{-1} · kg^{-1}, respectively), were 7–20 times as large as those observed in the current study (37 ml · min^{-1} · kg^{-1}). In addition, the elimination half-life of succinylcholine (16.6 and 11.7 s, respectively) was only 30% of that observed in our study. The discrepancy is probably the result of a lack of sensitivity of the analytical method in the previous study as succinylcholine could not be quantified more than 2 min after its administration. This would also explain the faster elimination half-life reported in that study.

Many methodologic issues had to be considered in the design of our study. To follow adequately the concentration–time profile immediately after the bolus injection, our study design included arterial sampling every 5 s for the first 2 min. The possibility of a random error in timing cannot be excluded but can be substantially reduced by strict adherence to the experimental protocol and by having the same persons responsible for blood sampling (experienced anesthetist) and time recording (experienced investigator). Any deviation from schedule was taken into account during the analysis. To minimize the error in the estimation of clearance, sampling from the arterial site was conducted from time of injection as frequently as possible.[21] In contrast to previous studies conducted by this group with neuromuscular blocking agents,[3,22,23] it was felt necessary to increase the frequency of blood sampling from every 10 s to every 5 s in view of the very fast onset of action of succinylcholine. A collection period of 10 min was judged adequate as the extrapolated terminal portion of the area under the curve was less than 2% of the total

Table 4. Pharmacokinetic–Pharmacodynamic Analysis

	Nonparametric Approach				Parametric Approach			
Patient No.	ke0 (min^{-1})	γ	EC_{50} (ng/ml)	AIC	ke0 (min^{-1})	γ	EC_{50} (ng/ml)	AIC
1	0.03066	8.56	640	377	0.0318	18.6	630	276
2	0.03738	26.64	549	196	0.03804	27.1	671	181
3	0.06252	11.25	850	236	0.06948	12.4	855	174
4	0.03324	7.55	420	312	0.0327	33.7	397	177
5	0.06174	8.61	743	270	0.06774	15.5	788	180
6	0.07614	4.22	953	303	0.07476	14.0	995	152
7	0.10254	6.65	985	262	0.09474	13.5	970	161
Mean	0.0577	10.5	734	279.3	0.0585	19.3	758	185.8
SD	0.0262	7.4	211	58.2	0.0244	8.1	210	41.3

AIC = Akaike information criterion.

area under the curve. Neuromuscular function, however, was monitored every 10 s only, in agreement with the recommendations for studies on neuromuscular blocking agents.[24] Therefore, effect was not measured as often as concentration during onset. The *in vitro* samples were incubated at the same temperature as that of the patient because temperature is known to influence the enzyme activity.[25]

As reported previously, the phenotyping results using conventional biochemical methods do not always allow correct genotyping or prediction of the response to neuromuscular blocking agents, and many patients cannot be classified using the traditional methods.[26,27] It was previously found that in 28.1% of 225 patients who presented an episode of prolonged apnea after succinylcholine administration, both the type and quantity of plasma cholinesterase were normal.[2] Subjects with genotypically normal enzyme but with low cholinesterase activity showed an increase in the duration of action of succinylcholine with decreasing cholinesterase activity.[28] In this study, the two patients with the lowest and highest cholinesterase activity (patients no. 1 and 4, respectively) also showed, respectively, the longest and the shortest recovery index, which is in agreement with previous findings. Two patients (no. 1 and 2) showed a relatively low plasma cholinesterase activity and had longer duration of action and plasma elimination half-life than the other subjects. In these patients, a lower cholinesterase activity was associated with EC_{50} values within the range observed in our patients (table 2). These results suggest that increased plasma concentrations, and not increased sensitivity, are probably responsible for the changes in pharmacodynamic response and the lower dose required in one case report of a homozygous patient having a atypical plasma cholinesterase.[29]

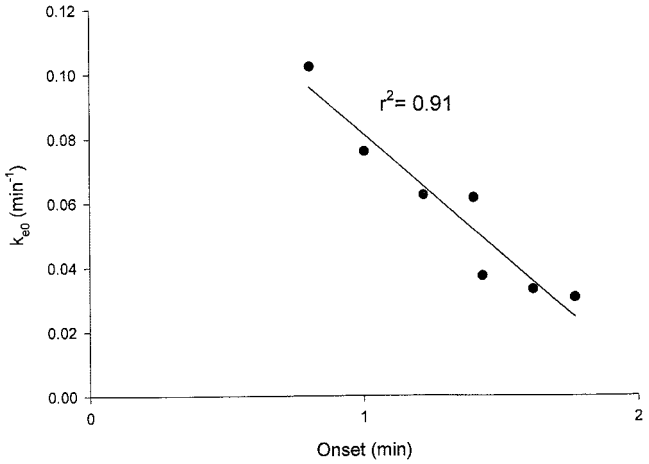

Fig. 6. Linear relation between the effect compartment equilibration rate constant (k_{e0}) and onset for seven patients using the noncompartmental pharmacokinetic–pharmacodynamic approach.

The advantage of measuring the *in vitro* rate of degradation of succinylcholine is its strong correlation with the *in vivo* rate of elimination in the same patient. Therefore, it would be interesting to investigate if the *in vitro* rate of degradation of succinylcholine could be a more robust method to detect, prior to anesthesia, the patients susceptible to have a prolonged apnea after succinylcholine administration, instead of conventional biochemical tests, as the latter do not seem to be good predictors. The determination of the *in vitro* rate of degradation of succinylcholine would be of clinical value in patients with a family history of prolonged apnea. However, quantitative determination of succinylcholine is not readily feasible in clinical practice.

As mentioned in Methods, we also believed that accounting for both central and peripheral elimination was appropriate for a more accurate estimation of the volume of distribution for succinylcholine ($V_{ss,c+p}$). The use of a $k_{in\ vitro}$ as a substitute for K_{20} was first proposed by Fisher *et al.*[30] for atracurium. For cisatracurium, the $k_{in\ vitro}$ value obtained by Welch *et al.*[31] had to be used, in the absence of the patient's *in vitro* data.[32–34] In our study, $k_{in\ vitro}$ values were obtained for six patients, but a large interpatient variability was observed. The use of this group of patient's mean $k_{in\ vitro}$ value would lead to significant error in the estimation of $V_{ss,c+p}$ in many of our patients. Recently, we proposed an alternative approach where k_{20} is assumed to be equal to β.[13] This

approach would be only indicated when the *in vivo* elimination half-life and the *in vitro* rate of degradation in plasma of a drug are similar in the same patient. We have therefore tested that assumption for succinylcholine and found that the $k_{in\ vitro}$ ($1.07 \pm 0.49\ min^{-1}$), k_{el} ($1.01 \pm 0.29\ min^{-1}$), and β ($1.01 \pm 0.28\ min^{-1}$) values were not statistically different for succinylcholine. In a model assuming both central and peripheral elimination,[13] it mathematically follows that the elimination rates in both compartments are identical. Although it is impossible to give a physiologic meaning to these compartments, our results suggest that the organ independent clearance (*e.g.*, hydrolysis by plasma cholinesterase) accounts for all of the total body clearance. Therefore, k_{20} could be fixed to β without the necessity of performing individual *in vitro* incubations in future studies involving succinylcholine.

When using compartmental models, it is assumed that the sampling site (the intravascular space) is part of a homogenous compartment (V_1), which may not be the case immediately after an intravenous bolus injection. Indeed, in one cycle of circulation, it would be unrealistic to presume that the central compartment is well stirred. Using physiologic markers, such as indocyanine green, allows full characterization of the intravascular mixing of muscle relaxants and has been shown to include not only the central circulation but also two peripheral circuits that return drug to the central compartment without equilibrating with tissues.[35] In our patients, it was possible to identify, by visual inspection of the initial front-end kinetics, a data point that could be associated with a small recirculation peak. This increase occurred after the initial mixing phase at approximately 65 ± 13 s, but its magnitude was within the analytical error range ($< 15\%$) in five of seven patients. As the initial intravascular mixing phase of succinylcholine was well characterized by the input function (k_{01}), we opted for a two-compartment pharmacokinetic model. To obtain a reasonable fit with experimental data, a lag time and an exponential input function k_{01} (analogous to an oral absorption function) had to be added to the two-compartment model to account for the intravascular mixing phase. In our study, the noncompartmental approach was used a model-independent reference to validate the compartmental model. The close concordance between both analyses provides additional support to our modeling approach.

Succinylcholine is expected to be very hydrosoluble and thus to have a small volume of distribution. The $V_{ss,c}$ derived with either the noncompartmental or compartmental analyses are similar to that reported by Hoshi *et al.*,[5] 19 or 20 ml/kg *versus* 16.4 ml/kg, respectively. Thus, the body distribution of succinylcholine would appear to be smaller than the intravascular space. However, it is generally recognized that the apparent V_{ss} of a drug will be underestimated if peripheral elimination is not taken into account.[13,30] After correction for peripheral elimination, the body distribution of succinylcholine appears to correspond to the intravascular space ($V_{ss,c+p} = 39$ ml/kg). In view of the extremely rapid clearance of succinylcholine, the hypothesis that a significant amount of the intravenous dose would be metabolized before its first detection in the arterial blood was investigated. The initial central volume of distribution for a series of muscle relaxants,[3,22,23] where extensive blood sampling was conducted during the first 2 min, were grossly estimated by dividing the dose by the maximum plasma concentrations. To our surprise, the volume obtained for succinylcholine was not that different from those having a slower plasma clearance. This observation would therefore exclude the possibility of a major contribution of the cardiopulmonary first-pass effect to the underestimation of the apparent volume of distribution of succinylcholine.

Everyone now agrees that it is of prime importance to characterize the front-end kinetics for a drug with a very rapid onset of action. Recently, a complete recirculatory model was used to describe the front-end kinetics of rocuronium in patients. In this study, blood samples were drawn at 3-s intervals during the first 2 min, and indocyanine green was used as a physiologic marker.[36] In their study, a smaller V_1 was reported for rocuronium when compared with that observed by our group using a 10-s sampling interval and noncompartmental analysis.[37] However, pharmacokinetic–pharmacodynamic estimates (k_{eo} and EC_{50} values) were similar in both studies. This suggests that the recirculation peaks have no significant impact on the estimation of pharmacokinetic–pharmacodynamic parameters.

When conducting nonparametric pharmacokinetic–pharmacodynamic modeling, some linear interpolation of the effect data had to be made during the first 2 min after succinylcholine administration. Also, because the effect lasted longer than plasma concentrations were quantifiable, the terminal portion of the concentration–time profile had to be extrapolated to get the corresponding concentration. The advantage of the parametric pharmacokinetic–pharmacodynamic analysis is that these interpolations or extrapolations are done automatically. Although the γ differed in both analysis, the estimates of EC_{50} and k_{e0} were similar.

Transient muscular fasciculations were observed in two patients, but no significant twitch increase was seen on the twitch tracing. Thus, onset of action could be considered as unimodal and was treated as such. Recovery occurred much earlier than the time required for phase II block to develop (30–45 min). Thus, the changing nature of succinylcholine action did not have to be taken into consideration in our pharmacodynamic model, and the traditional sigmoid E_{max} model was applied to our data. Our results indicate that the potency of succinylcholine is low. Indeed, its molar EC_{50} (2.6 μM) is almost 5-fold that of atracurium (0.49 μM), 21-fold that of

doxacurium (0.12 μM), and 47-fold that reported for mivacurium (0.055 μM).[22,38,39]

The k_{e0} is thought to be governed by several factors, such as perfusion to the effect site, diffusion from the capillary lumen to the effect site (molecular weight, pKa, lipid solubility, protein binding), and occupancy of receptors to induce drug effect (potency).[40,41] Interestingly, a strong correlation between k_{e0} and onset was observed in our patients ($r^2 = 0.9$) (fig. 6). Despite its fast onset of action, the equilibration half-life between succinylcholine plasma concentrations and its effect was relatively slow, being similar to that obtained for various nondepolarizing neuromuscular blocking agents; as such, mean k_{e0} values of 0.043 ± 0.004 min^{-1} were reported for atracurium, 0.053 ± 0.006 min^{-1} for doxacurium, and 0.058 ± 0.005 min^{-1} for vecuronium.[3,22,42] Recently, Torda *et al.*[43] used a pharmacodynamic model to derive the pharmacokinetic–pharmacodynamic parameters for succinylcholine from effect data alone and found a k_{e0} of 0.27 ± 0.15 min^{-1}, which is almost five times as fast as what we obtained. The fact that plasma concentrations of succinylcholine were not measured in their patients may well explain these discrepancies. In a study involving rapacuronium, a nondepolarizing blocking agent with a fast onset of action, a k_{e0} ranging from 0.377 to 0.405 min^{-1} was reported at the adductor pollicis.[44] It was concluded that the more rapid onset of rapacuronium compared with the other nondepolarizing muscle relaxants resulted from a more rapid equilibration between plasma and effect site concentrations.[44] The same conclusion cannot be applied to succinylcholine. Pharmacokinetic factors other than a fast transfer to the site of action are probably responsible for the rapid onset of action of succinylcholine, namely, the combination of a low potency and high plasma clearance.[45,46]

In conclusion, succinylcholine is a low potency drug with very fast clearance that equilibrates relatively slowly with the effect compartment. Its very fast disappearance from plasma has been characterized *in vivo* and *in vitro*, and the *in vitro* speed of degradation is a strong predictor of the *in vivo* elimination rate constant.

The authors thank Orval A. Mamer, Ph.D. (Mass Spectrometry Unit, McGill University), for his sustained support, and Denis Babin, M.Sc. (Centre Hospitalier de l'Université de Montréal), Johanne Couture, R.T., and Nathalie Rivest, B.Pharm. (Faculté de Pharmacie, Université de Montréal) for their collaboration in this study.

References

1. Whittaker V, Wijesundera S: The hydrolysis of succinylcholine by cholinesterase. Biochem J 1952; 52:475–9
2. Viby-Mogensen J, Hanel HK: Prolonged apnoea after suxamethonium: An analysis of the first 225 cases reported to the Danish Cholinesterase Research Unit. Acta Anaesthesiol Scand 1978; 22:371–80
3. Ducharme J, Varin F, Bevan DR, Donati F: Importance of early blood sampling on vecuronium pharmacokinetic and pharmacodynamic parameters. Clin Pharmacokinet 1993; 24:507–18
4. Runciman WB, Upton RN: Pharmacokinetics and pharmacodynamics: What is of value to the practising anaesthetist? Anaesthetic pharmacol Rev 1994; 2:280–93
5. Hoshi K, Hashimoto Y, Matsukawa S: Pharmacokinetics of succinylcholine in man. Tohuku J Exp Med 1993; 170:245–50
6. Roy JJ, Boismenu D, Gao H, Mamer OA, Varin F: Measurement of succinylcholine concentration in human plasma by electrospray tandem mass spectrometry. Anal Biochem 2001; 290:238–45
7. Koelle G, Steiner E: The cerebral distributions of a tertiary and a quaternary anticholinesterase agent following intravenous and intraventricular injection. J Pharmacol Exp Ther 1956; 118:420–34
8. Longpre J: [Qualitative and quantitative measurement of the activity of pseudocholinesterases]. Can J Med Technol 1971; 33:213–27
9. Hartley HO: The modified Gauss-Newton method for fitting of non-linear regression functions by least squares. Technometrics 1961; 3:269–80
10. Davies M, Whitting IJ: A modified form of Levenberg's correction, Numerical Methods for Non-Linear Optimization. Edited by Lootsma FA. London, Academic Press, 1972, pp 191–201
11. Gibaldi M, Perrier D: Pharmacokinetics. Edited by Gibaldi M, Perrier D. New York, Marcel Dekker, 1982, pp 1–494
12. Nakashima E, Benet LZ: General treatment of mean residence time, clearance, and volume parameters in linear mammillary models with elimination from any compartment. J Pharmacokinet Biopharm 1988; 16:475–92
13. Laurin J, Nekka F, Donati F, Varin F: Assuming peripheral elimination: Its impact on the estimation of pharmacokinetic parameters of muscle relaxants. J Pharmacokinet Biopharm 1999; 27:491–512
14. Yamaoka K, Nakagawa T, Uno T: Application of Akaike's information criterion (AIC) in the evaluation of linear pharmacokinetic equations. J Pharmacokinet Biopharm 1978; 6:165–75
15. Ezzine S, Donati F, Varin F: Aterial-venous gradient under mivacurium steady-state concentrations in anesthetized patients (abstract). ANESTHESIOLOGY 2001; 95:A468
16. Jacquez JA: Compartmental Analysis in Biology and Medicine. Edited by Jacquez JA. Ann Arbor, BioMedware, 1996, pp 1–514
17. Unadkat JD, Bartha F, Sheiner LB: Simultaneous modeling of pharmacokinetics and pharmacodynamics with nonparametric kinetic and dynamic models. Clin Pharmacol Ther 1986; 40:86–93
18. Sheiner LB, Stanski DR, Vozeh S, Miller RD, Ham J: Simultaneous modeling of pharmacokinetics and pharmacodynamics: Application to d-tubocurarine. Clin Pharmacol Ther 1979; 25:358–71
19. Fuseau E, Sheiner LB: Simultaneous modeling of pharmacokinetics and pharmacodynamics with a nonparametric pharmacodynamic model. Clin Pharmacol Ther 1984; 35:733–41
20. Holford NH, Sheiner LB: Understanding the dose-effect relationship: Clinical application of pharmacokinetic-pharmacodynamic models. Clin Pharmacokinet 1981; 6:429–53
21. Weiss M: Errors in clearance estimation after bolus injection and arterial sampling: Nonexistence of a central compartment. J Pharmacokinet Biopharm 1997; 2:255–60
22. Zhu Y, Audibert G, Donati F, Varin F: Pharmacokinetic pharmacodynamic modeling of doxacurium: Effect of input rate. J Pharmacokinet Biopharm 1997; 25:23–37
23. Lacroix M, Donati F, Varin F: Pharmacokinetics of mivacurium isomers and their metabolites in healty volunteers after intravenous bolus administration. ANESTHESIOLOGY 1997; 86:322–30
24. Viby-Mogensen J, Engbaek J, Eriksson LI, Gramstad L, Jensen E, Jensen FS, Koscielniak-Nielsen Z, Skovgaard LT, Ostergaard D: Good clinical research practice (GCRP) in pharmacodynamic studies of neuromuscular blocking agents. Acta Anaesthesiol Scand 1996; 40:59–74
25. Foldes FF: Enzymes of acetylcholine metabolism, Enzymes in Anesthesiology. Edited by Foldes FF. New York, Springer-Verlag, 1978, pp 91–168
26. Rosenberg MK, Lebenbom-Mansour M: Markedly prolonged paralysis after mivacurium in a patient apparently heterozygous for the atypical and usual pseudocholinesterase alleles by conventional biochemical testing. Anesth Analg 1997; 84:457–60
27. Jensen FS, Skovgaard LT, Viby-Mogensen J: Identification of human plasma cholinesterase variants in 6,688 individuals using biochemical analysis. Acta Anaesthesiol Scand 1995; 39:157–62
28. Viby-Mogensen J: Correlation of succinylcholine duration of action with plasma cholinesterase activity in subjects with the genotypically normal enzyme. ANESTHESIOLOGY 1980; 53:517–20
29. Hickey DR, O'Connor JP, Donati F: Comparison of atracurium and succinylcholine for electroconvulsive therapy in a patient with atypical plasma cholinesterase. Can J Anaesth 1987; 34:280–3
30. Fisher DM, Canfell PC, Fahey MR, Rosen JI, Rupp SM, Sheiner LB, Miller RD: Elimination of atracurium in humans: Contribution of hofman elimination and ester hydrolysis versus organ-based elimination. ANESTHESIOLOGY 1986; 65: 6–12
31. Welch RM, Brown A, Ravitch J, Dahl R: The in vitro degradation of cisatracurium, the R, cis-R'-isomer of atracurium, in human and rat plasma. Clin Pharmacol Ther 1995; 58:132–42

32. Kisor DF, Schmith VD, Wargin WA, Lien CA, Ornstein E, Cook DR: Importance of the organ-independant elimination of cisatracurium. Anesth Analg 1996; 83:1065–71

33. Bergeron L, Bevan DR, Berrill A, Kahwaji R, Varin F: Concentration-effect relationship of cisatracurium at three different dose levels in the anesthetized patient. ANESTHESIOLOGY 2001; 95:314–23

34. Lien CA, Schmith VD, Belmont MR, Abalos A, Kisor DF, Savarese JJ: Pharmacokinetics of cisatracurium in patients receiving nitrous oxide/opioid/barbiturate anesthesia. ANESTHESIOLOGY 1996; 84:300–8

35. Krejcie TC, Henthorn TK, Niemann CU, Klein C, Gupta DK, Brooks Gentry W, Shanks CA, Avram MJ: Recirculatory pharmacokinetic models of markers of blood, extracellular fluid and total body water administered concomitanly. J Pharmacol Exp Ther 1996; 278:1050–7

36. Kuipers JA, Boer F, Olofsen E, Bovill JG, Burm AG: Recirculatory pharmacokinetics and pharmacodynamics of rocuronium in patients. ANESTHESIOLOGY 2001; 94:47–55

37. Dragne A, Varin F, Plaud B, Donati F: Rocuronium pharmacokinetic-pharmacodynamic relationship under propofol and isoflurane anesthesia. Can J Anaesth 2002; 49:353–60

38. Donati F, Gill SS, Bevan DR, Ducharme J, Theoret Y, Varin F: Pharmacokinetics and pharmacodynamics of atracurium with and without previous suxamethonium administration. Br J Anaesth 1991; 66:557–61

39. Laurin J, Donati F, Nekka F, Varin F: Peripheral link model as an alternative for pharmacokinetic-pharmacodynamic modeling of drugs having a very short elimination half-life. J Pharmacokinet Biopharm 2001; 28:7–25

40. Hennis PJ, Stanski DR: Pharmacokinetic and pharmacodynamic factors that govern the clinical use of muscle relaxants. Semin Anesth 1985; 4:21–30

41. Stanski DR, Ham J, Miller RD, Sheiner LB: Pharmacokinetics and pharmacodynamics of d-tubocurarine during nitrous oxide-narcotic and halothane anesthesia in man. ANESTHESIOLOGY 1979; 51:235–41

42. Ducharme J, Varin F, Donati F: Pharmacokinetics and pharmacodynamics of a second dose of atracurium in anaesthetised patients. Clin Drug Invest 1995; 9:98–110

43. Torda T, Graham G, Warwick N, Donohue P: Pharmacokinetics and pharmacodynamics of suxemethonium. Anaesth Intensive Care 1997; 25:272–8

44. Wright PM, Brown R, Lau M, Fisher DM: A pharmacodynamic explanation for the rapid onset/offset of rapacuronium bromide. ANESTHESIOLOGY 1999; 90:16–23

45. Beaufort TM, Nigrovic V, Proost JH, Houwertjes MC, Wierda JM: Inhibition of the enzymic degradation of suxamethonium and mivacurium increases the onset time of submaximal neuromuscular block. ANESTHESIOLOGY 1998; 89:707–14

46. Kopman AF, Klewicka MM, Kopman DJ, Neuman GG: Molar potency is predictive of the speed of onset of neuromuscular block for agents of intermediate, short, and ultrashort duration. ANESTHESIOLOGY 1999; 90:425–31