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Pharmacokinetics and Pharmacodynamics of Mivacurium in Patients Phenotypically Homozygous for the Atypical Plasma Cholinesterase Variant

Effect of Injection of Human Cholinesterase

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Background: **In patients homozygous for atypical plasma cholinesterase, mivacurium causes a long-lasting neuromuscular block, but injection of human cholinesterase has been proven effective in antagonizing the block. The purpose of this study was to evaluate the pharmacodynamics and pharmacokinetics of mivacurium in such patients, as well as the effect of cholinesterase injected early or late after mivacurium.**

Methods: **Eleven patients phenotypically homozygous for the atypical variant received 0.075 mg/kg (1 patient) or 0.15 mg/kg (10 patients) mivacurium. The neuromuscular block was monitored using train-of-four nerve stimulation and mechanomyography. Cholinesterase, 2.8–10.0 mg/kg, was administered approximately 30 or 120 min after mivacurium. The times to different levels of neuromuscular recovery and the venous concentrations of the isomers of mivacurium were measured.**

Results: **Injection of cholinesterase increased plasma cholinesterase activity to normal and the clearances of the active isomers and the elimination rate constants by a factor of 10–15. The first response was seen in 13.5 min (3.7–44.2 min). Time to a train-of-four ratio of 0.8 ranged from 30 to 60 min (n = 6). Neostigmine injected after cholinesterase shortened recovery further, and a train-of-four ratio of 0.8 was reached in 10–30 min.**

Conclusion: **As expected, the duration of action of mivacurium is markedly prolonged in homozygous atypical patients. Injection of cholinesterase significantly increases the metabolism of mivacurium, leading to a shorter duration of action. Injection of neostigmine after the administration of cholinesterase speeds up recovery.**

IN patients with normal plasma cholinesterase (pChe) activity and phenotype, mivacurium is rapidly hydrolyzed by pChe.[1,2] In patients phenotypically homozygous for atypical pChe, mivacurium is approximately 5 times more potent than in normal patients.[3] In accordance with this, a small dose of mivacurium (0.03 mg/kg) causes 100% block, and the time to reappearance of the first response to train-of-four stimulation (TOF) has been found to vary from 26–128 min in five patients with this phenotype.[4] Further, several case reports describes very prolonged duration of action with 6–8 h to full recovery after a normal tracheal intubation dose of mivacurium (0.15–0.20 mg/kg).[5–7] Preliminary studies indicate that injection of purified human serum cholinesterase may be effective in reversing the neuromuscular block.[8,9] However, the optimal dose and optimal time of injection is not known, nor is the pharmacokinetic profile of mivacurium in these patients.

The purpose of the current study was to evaluate the pharmacodynamics and pharmacokinetics of mivacurium in patients phenotypically homozygous for the atypical pChe variant and further to evaluate the effects of various doses of human cholinesterase injected early (30 min) and late (120 min) after the administration of mivacurium on the speed of recovery.

Materials and Methods

Eleven patients with American Society of Anesthesiologists physical status I or II and phenotypically homozygous occurrence of the atypical gene who were scheduled to undergo elective surgery were enrolled in the study during a period of 5 yr. All patients had previously been issued warning cards by the Danish Cholinesterase Research Unit.[10] Patients with a history of neuromuscular, cardiovascular, renal, or hepatic disorders were excluded from the study, as were females of childbearing potential and patients receiving drugs that might affect neuromuscular transmission. The patients gave informed consent, and the Ethics Committee in Copenhagen County, Denmark, approved the study.

Anesthesia

Premedication consisted of 0.1–0.2 mg/kg oral diazepam. The actual pChe activity was determined from a blood sample taken before induction of anesthesia[11] (table 1). Anesthesia was induced with 1–4 μg/kg fentanyl, 0.05–0.15 mg/kg diazepam, and 3–6 mg/kg thiopentone and maintained with 66% nitrous oxide in oxygen and supplementary doses of fentanyl, and in a few patients, with 5–10 mg · kg^{-1} · h^{-1} propofol.

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Table 1. Biochemical Characteristics of the 11 Patients before and after Administration of Human Cholinesterase

Patient No.	Plasma Cholinesterase Activity Preoperatively, U/l	Dibucaine No.	Dose of Human Cholinesterase Injected, mg/kg	Plasma Cholinesterase Activity after Injection, U/l	Efficient Plasma Cholinesterase Activity, U/l
1	183	21	5.0	1,108	925
2	198	27	4.0	1,326	1,128
3	342	23	5.2	1,342	1,000
4	339	14	10.0	2,271	1,932
5	149	27	7.1	1,909	1,760
6	465	28	9.8	1,902	1,437
7	354	26	4.1	1,167	813
8	301	27	4.6	887	586
9	328	19	4.4	947	619
10	402	28	5.5	1,630	1,228
11	369	28	6.6	1,376	1,007

Reference values of the Danish Cholinesterase Research Unit for plasma cholinesterase activity and dibucaine number are 660–1,620 U/l and 79–87, respectively, for phenotypically normal patients, and 169–709 U/l and 13–28, respectively, for homozygous patients.[11] Efficient pChe = $pChe_{after} - pChe_{before}$, where $pChe_{after}$ is the activity measured after the injection of human cholinesterase, and $pChe_{before}$ is the activity before the injection.

During surgery, the patients were monitored continuously using electrocardiography, pulse oximetry, and capnography. Blood pressure was measured every minute for the first 3 min after administration of mivacurium and every 5 min thereafter. The blood pressure cuff and the intravenous line used for administration of anesthetics and fluid were on the same arm. Ventilation was adjusted to maintain normocapnia (end-tidal carbon dioxide pressure 34–42 mmHg). The rectal and peripheral skin temperatures were measured and maintained above 35° and 32°C, respectively.[12] After induction of anesthesia, a second intravenous catheter was inserted for blood sampling in the arm used for monitoring.

At the start of the study, neither the duration of action of normal tracheal intubation doses of mivacurium nor the effectiveness of cholinesterase were known in these patients. Therefore, the first patient received 0.075 mg/kg, and the remaining 10 patients (patients 2–11) were given 0.15 mg/kg mivacurium, which represents a clinically relevant dose for induction of anesthesia.

To evaluate the effect of mivacurium after the injection of cholinesterase and after recovery of neuromuscular block following the first dose of mivacurium, one patient (patient 4) received two supplementary doses of 0.03 mg/kg mivacurium, and another patient (patient 5) received 0.03 and 0.06 mg/kg mivacurium.

Neuromuscular Monitoring

The mechanical twitch was recorded using a Myograph 2000 (Biometer International, Odense, Denmark). The ulnar nerve was stimulated at the wrist using surface electrodes and 1-Hz single-twitch stimulation. After supramaximal stimulation was achieved, the stimulation pattern was changed to TOF stimulation every 12 s. When the response to stimulation was stable for 5 min, a single bolus dose of mivacurium was given over 30 s.

Tracheal intubation was performed at maximum T_1 suppression, where T_1 is the first response in the TOF response.

Onset (time from beginning of injection of mivacurium to 95% T_1 suppression) and recovery data were determined using start control values, and monitoring was continued at least until 90% T_1 recovery and a TOF ratio of 0.80 were obtained.[12] The time to first response to TOF stimulation and the durations to 10, 25, 50, and 90% T_1 recovery and a TOF ratio of 0.80 were determined. The interval from 25% to 75% twitch height recovery was also calculated. In patients given neostigmine for reversal of neuromuscular block (see Injection of Neostigmine), the times from administration of neostigmine to 90% T_1 recovery and to a TOF ratio of 0.80 were measured.[12]

Injection of Human Cholinesterase

We used serum cholinesterase, Centeon® (Centeon Pharma, Marburg, Germany), a highly purified concentrate of the enzyme. The content of each vial, 55 mg (27–83 mg), is equivalent in activity to 500 ml fresh human plasma.

Six patients with expected short-lasting surgeries received cholinesterase 30–36 min after the injection of mivacurium (patients 1–6), and five patients with expected long-lasting surgeries received cholinesterase after 82–143 min (patients 7–11).

In the first seven patients given cholinesterase, a dose of 4.0–5.5 mg/kg was administered (patients 1, 2, 3, 7, 8, 9, and 10; table 1), doses that increased the effective enzyme activity to within normal levels in all patients. To evaluate whether an even higher dose of cholinesterase would further increase the metabolic rate of mivacurium and thus shorten the recovery, four patients (patients 4, 5, 6, and 11; table 1) received 6.6–10.0 mg/kg cholinesterase.

Measurement of Plasma Cholinesterase Activity and Determination of Phenotypes

Plasma cholinesterase activity was measured using benzoylcholine as a substrate, and the previously found

phenotype was confirmed using several inhibitors.[11] However, only the results regarding the dibucaine number are given.

Blood samples for measurement of pChe activity were collected immediately before induction of anesthesia, before and 5–15 min after administration of human cholinesterase, and before and 5 min after administration of neostigmine. In four patients, samples for measurement of pChe were also collected 30 and 60 min after administration of human cholinesterase. The amount of active (or efficient) enzyme ($pChe_{efficient}$) in blood was calculated as: $pChe_{efficient} = pChe_{after} - pChe_{before}$, where $pChe_{after}$ is the activity measured after the injection of human cholinesterase, and $pChe_{before}$ is the activity before the injection.

Injection of Neostigmine

In principle, neuromuscular block was left to recover "spontaneously" after injection of cholinesterase (patients 1, 3, 4, 6, 9, and 10). However, when surgery ended before sufficient recovery of neuromuscular function after injection of cholinesterase was achieved, 0.04 mg/kg neostigmine preceded by atropine was given (patients 2, 5, 7, 8, and 11).

Plasma Concentrations of Mivacurium

Venous blood samples (5 ml) were collected immediately before administration of mivacurium; 1, 2, 5, 10, 15, 20, 30, and 60 min after the start of injection; and 1, 2, 5, 10, 15, 20, 30, 60, and 120 min after injection of human cholinesterase.[13] In less than 10 s, the blood was transferred into a vacutainer containing a cholinesterase inhibitor (phospholine iodide). The samples were centrifuged, and the plasma was separated and frozen at −70°C. The ratio of cis-cis, cis-trans, and trans-trans isomers in the clinical trial material used were approximately 5.8, 35.5, and 58.8%, respectively (data from certificate of analysis, GlaxoSmithKline [formerly GlaxoWellcome, Beckenham, United Kingdom]). The concentration of each isomer of mivacurium was determined by a stereospecific high-performance liquid chromatographic method with fluorometric detection and a stepped gradient. The coefficient of variation was less than 10% at all concentrations except for the lowest level of quantification (5 ng/ml) (14%). Extraction efficiency was 75%. Calibration was linear over the range 5–1,000 ng/ml.[13] The relative error was less than 6% for the isomers.

Pharmacokinetics

A two-compartment model was fitted to the plasma concentrations of each isomer separately. The fitting was performed using numerical integration of differential equations and using the inverse square of the calculated plasma concentration of mivacurium as a weight factor.[14] After the administration of cholinesterase, an extra k'_e was numerically added to the elimination rate constant. The parameters V_1, k_e, k_{12}, k_{21}, and k'_e were fitted to the data, and the fitting for the elimination rate constant K was as follows: before cholinesterase $K = k_e$; after cholinesterase $K = k_e + k'_e$. The extra k'_e was removed at the time of the administration of neostigmine. The goodness of fit was evaluated by visual inspection and from plots of residuals. The following secondary parameters were calculated before and after administration of cholinesterase: clearance, $CL = V_1 * K$; volume of distribution at steady state, $V_{ss} = V_1 * (k_{21} + k_{12})/k_{21}$; and the terminal rate constant λ_2 in the two-compartment model was calculated from the primary parameters as

$$\lambda_2 = \tfrac{1}{2}\left[k_{12} + k_{21} + K - \sqrt{(k_{12} + k_{21} + K)^2 - 4k_{21}K} \right].$$

Table 2. Recovery Data after Injection of Human Cholinesterase in Patients Phenotypically Homozygous for the Atypical Plasma Cholinesterase Enzyme Given Mivacurium and Cholinesterase

Patient No.	Dose of Mivacurium, mg/kg	Time to Injection of Cholinesterase, min	Time to T_1 after Injection of Cholinesterase, min	Duration, min				25–75% Interval, min	Duration TOF 0.80, min
				10%	25%	50%	90%		
1	0.075	31	14.5	14.7	18.0	21.3	30.0	7.7	32.0
2	0.150	36	35.5	46.1*	—	—	—	—	—
3	0.150	33	11.7	20.2	25.3	32.8	—	—	52.2
4	0.150	32	13.3	18.1	23.4	31.5†	—	18.0	—
5	0.150	30	44.2	56.7	71.8*†	—	—	—	—
6	0.150	31	21.7	27.3	32.0	39.7	64.0	19.6	59.3
7	0.150	82	13.9	19.6*	—	—	—	—	—
8	0.150	114	3.7	8.8	14.2*	—	—	—	—
9	0.150	113	10.2	12.9	15.9	20.0	26.0	7.4	29.8
10	0.150	143	8.2	13.4	17.4	22.6	37.0	12.3	41.4
11	0.150	117	14.7*	—	—	—	—	—	—

Values given are minutes. Data are presented according to the Good Clinical Research Practice rules for pharmacodynamic studies in neuromuscular blocking agents.[12]

* Patients later given neostigmine. † Indicates patients given a supplementary dose of mivacurium (recovery data not shown).

T_1 = time to reappearance of the first response to TOF stimulation after the injection of cholinesterase; TOF = train-of-four ratio.

Table 3. Recovery Data after Injection of Neostigmine (Administered after Human Plasma Cholinesterase

Patient No.	Time after Cholinesterase, min	T_1 before Injection, %	Duration, min				25–75% Interval, min
			25%	50%	90%	TOF 0.80	
2	49	15	1.9	4.6	25.7	29.3	7.5
7	22	18	1.0	2.0	7.3	12.1	4.2
8	16	31	—	1.0	4.2	10.0	—
11	20	8	2.2	7.2	15.7	13.7	9.3

Values given are minutes.

T_1 = time to reappearance of the first response to TOF stimulation; TOF = train-of-four stimulation.

The clearance of each isomer was calculated from the time of injection of mivacurium until the administration of cholinesterase. The clearance after the injection of human cholinesterase was calculated from the time of the injection until infinity.

Pharmacokinetic–Pharmacodynamic Modeling

Pharmacokinetic–pharmacodynamic modeling was performed in two steps. The first step was fitting the described pharmacokinetic model to concentrations calculated as concentration of trans-trans isomer plus 0.9 concentration of the cis-trans isomer. The contribution of the less potent cis-cis isomer was ignored. Second, a link model between the effect and the central compartment concentrations was fitted to the effect data using fixed parameters for the pharmacokinetic part of the model. The sigmoid Emax model was fitted to the effect compartment concentrations: $E = (E_{max} \cdot C^{\gamma}_e)/EC^{\gamma}_{50} + C^{\gamma}_e$, where E is the intensity of pharmacologic effect (percent depression of T_1), Emax is the maximum effect, C_e is the concentration in the hypothetical effect compartment in the k_{eo} model described by Holford and Sheiner,[15] EC_{50} is the plasma concentration at 50% T_1 depression, and γ (the Hill coefficient) is the parameter determining the sigmoidicity of the relation between concentration and effect. Fitting and evaluation of the goodness of the fit were performed as described in the Pharmacokinetics section.

Statistical Analysis

All pharmacokinetic and pharmacodynamic results are presented as median and range. Data before and after administration of cholinesterase were compared using the nonparametric Mann–Whitney U test. $P < 0.05$ was considered significant.

Results

Two males and nine females were included in the study. The median age was 48 yr (range, 15–73 yr). All patients except one were within 20% of their ideal body weight (patient 3 was 30% above the ideal body weight), body weight being 65 kg (55–100 kg) and height being 167 cm (156–176 cm).

Enzyme Activity in Plasma before and after Injection of Cholinesterase

All patients had dibucaine numbers below 30 at baseline (table 1). After injection of human cholinesterase, pChe activity increased within 2–10 min in a dose-dependent manner.

Pharmacodynamics

Neuromuscular Blocking Effect of Mivacurium before Injection of Cholinesterase. Median onset times after 0.075 and 0.15 mg/kg mivacurium were 1.5 and 2.0 min (range, 1.0–2.3 min), respectively. The neuromuscular block was very deep at the time of injection of cholinesterase as indicated by no reaction to TOF.

Recovery of Neuromuscular Block after Injection of Cholinesterase. Table 2 shows times the recovery data after injection of cholinesterase. In all patients, the administration speeded up recovery. No significant rela-

Table 4. Duration of Action and Recovery Data after Supplementary Doses of Mivacurium Administered after Patients Received Human Plasma Cholinesterase

Patient No.	Supplementary Dose of Mivacurium, mg/kg	T_1 at time of Injection, %	Maximum Block, %	Time to T_1, min	Duration, min				25–75% Interval, min
					10%	25%	90%	TOF 0.80	
4	0.03	78	98	—	4.2	6.7	—	20.7	12.3
	0.03	80	98	—	4.8	7.1	19.6	18.6	9.2
5	0.03	35	100	5.16	14.0	19.8	51.6	44.7	15.3
	0.06	92	100	19.0	28.6	35.9*	—	—	—

Values given are minutes.

* Patient later given neostigmine (recovery data not shown).

T_1 = time to reappearance of the first response to TOF stimulation; TOF = train-of-four stimulation.

tion was found between the dose of injected cholinesterase and the rate of recovery. For example, the median times to reappearance of T_1 after the low and high doses of cholinesterase were 12.8 min (3.7–35.5 min) and 18.2 min (13.3–44.2 min), respectively (not significant).

With respect to the timing of the administration of cholinesterase, the data indicate a faster recovery of neuromuscular function after the late injection (patients 7–11) as compared with the early injection (patients 2–6). The median times to reappearance of T_1 and to T_1 10%, respectively, were 10.2 min (3.7–14.7 min) and 13.2 min (8.8–19.6 min) after the late injection, as compared with 21.7 min (11.7–44.2 min; not significant) and 27.3 min (18.1–56.7 min; $P < 0.05$) after the early injection of cholinesterase.

Recovery of Neuromuscular Block after Injection of Neostigmine. Table 3 shows recovery data following injection of neostigmine administered after cholinesterase. The injection of neostigmine caused a fast recovery in three of four patients. The 25–75% interval was 7.5 min (4.0–9.3 min), which was shorter than in patients who did not receive neostigmine 15.1 min (7.4–19.6 min). In one patient (patient 2), recoveries after injection of cholinesterase and after neostigmine were both very slow (table 3). One patient (patient 5) received neostigmine after the second supplementary dose of mivacurium at a T_1 of 42%, and a TOF ratio of 0.8 was obtained in 6 min.

Significance of Injected Cholinesterase for the Reaction to Supplementary Doses of Mivacurium. Recovery data following injection of the two supplementary doses of mivacurium after administration of cholinesterase (patients 4 and 5) are presented in table 4.

Pharmacokinetics

Pharmacokinetics of Mivacurium before Injection of Cholinesterase. Figure 1 shows the individual plasma concentrations of each of the three isomers over time after late injection of cholinesterase and neostigmine in one of the patients. The pharmacokinetic data for the three isomers are summarized in table 5. The clearances of all isomers and the elimination rate constants were decreased as compared with phenotypically normal patients indicating longer elimination half-lives (data from a previous study in phenotypically normal patients is presented in table 5 for comparison).[16] The clearances and the rate constants of the active isomers were increased more than the cis-cis isomer. The volume of distribution was small for all isomers as in phenotypically normal patients.

Pharmacokinetics of Mivacurium after Injection of Cholinesterase. Immediately after injection of cholinesterase, the concentration of the active isomers decreased rapidly, whereas the concentration of the cis-cis isomer was less affected (fig. 1). After administration of neostigmine, the concentration of the active isomers increased moderately (fig. 1).

The clearances and the elimination rate constants of all three isomers increased by a factor of approximately 10. The half-lives decreased correspondingly.

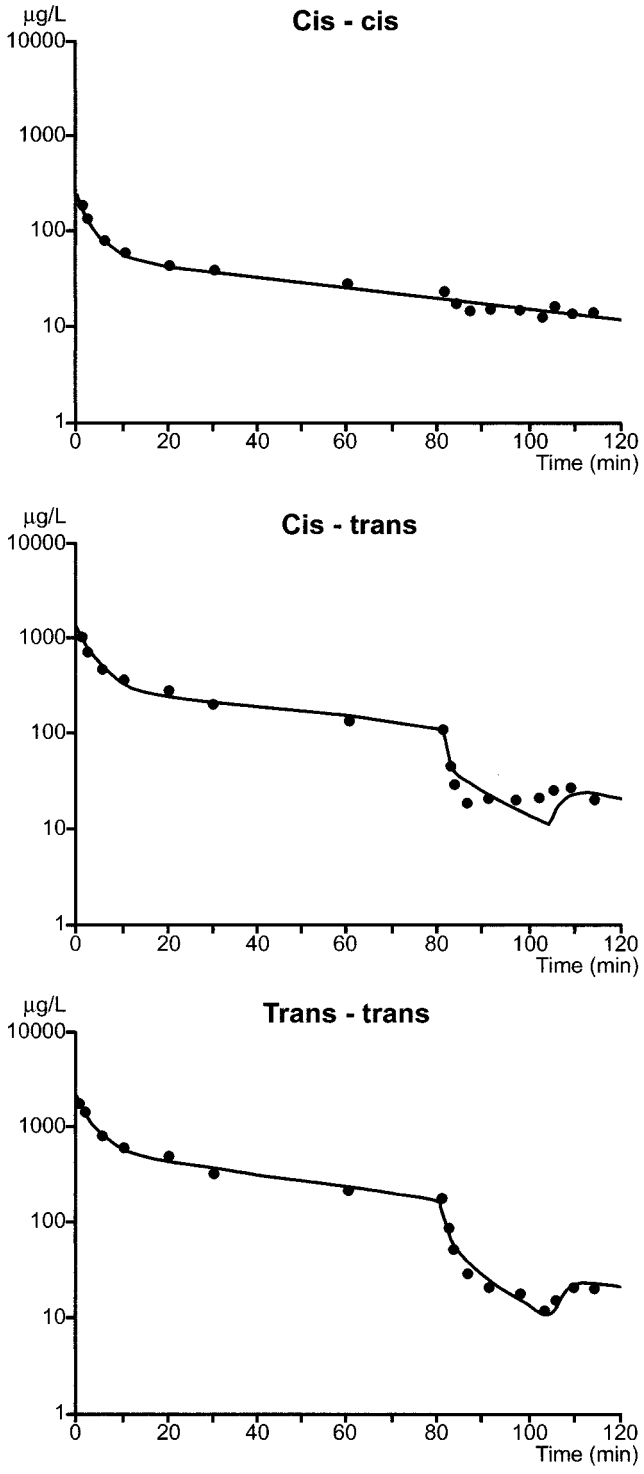

Fig. 1. Plasma concentrations (ng/ml) of the cis-cis, cis-trans, and trans-trans isomers over time (minutes) in phenotypically homozygous patient 7. Note that cholinesterase and neostigmine were administered 80 and 105 min after mivacurium, respectively.

Table 5. Estimated Clearance, Rate Constants k_e, k_{12}, and k_{21}, Elimination Half-life ($T_{1/2}{}^{\gamma}{}_z$) and Initial Volume of Distribution (V_1) in Patients Phenotypically Homozygous for the Atypical Plasma Cholinesterase Enzyme before and after Administration of Human Plasma Cholinesterase

Isomer	Clearance, ml · kg^{-1} · min^{-1}	k_e, min^{-1}	k_{12}, min^{-1}	k_{21}, min^{-1}	$T_{1/2}\lambda_z$, min	V_1, l/kg
Cis-cis						
Before	1.5 (0–12.9)	0.040 (0–0.176)	0.129 (0.052–0.327)	0.039 (0.005–0.305)	91.9 (50.4–133.3)	0.07 (0.02–0.34)
After	18.0 (1.2–153.0)	0.162* (0–2.03)			26.3* (3.3–133.1)	
Normal patients	6.0 (4.2–9.8)	0.110 (0.07–0.203)	0.110 (0.042–0.224)	0.077 (0.053–0.101)	22.6 (16.9–38.4)	0.05 (0.02–0.14)
Cis-trans						
Before	2.3 (0–5.7)	0.052 (0–0.388)	0.229 (0.066–0.723)	0.068 (0.026–0.297)	63.5 (53.3–116.1)	0.04 (0.01–0.18)
After	33.5* (8.9–258.1)	0.957* (0.215–4.559)			11.5* (2.7–27.1)	
Normal patients	50.3 (36.8–175.5)	0.647 (0.372–1.226)	0.218 (0.066–0.376)	0.147 (0.060–0.210)	6.7 (5.4–12.7)	0.08 (0.04–0.47)
Trans-trans						
Before	2.7 (0–5.1)	0.056 (0–0.093)	0.203 (0.023–0.680)	0.092 (0.030–0.310)	66.9 (38.8–79.5)	0.05 (0.02–0.14)
After	25.4* (8.2–122.5)	0.464* (0.293–2.448)			13.9* (3.1–23.7)	
Normal patients	27.6 (22.2–96.0)	0.635 (0.328–1.028)	0.195 (0.049–0.469)	0.149 (0.077–0.273)	7.2 (3.8–11.8)	0.05 (0.02–0.29)

$T_{1/2}\lambda_z$ before: only data from patients receiving cholinesterase late are given. For comparison, values for phenotypically normal patients are given.[16] Median and range are given.

* Statistically significant difference ($P < 0.05$).

Pharmacokinetic–Pharmacodynamic Modeling

Figure 2 illustrates the relation between the estimated effect compartment concentration and time. The observed plasma concentration decreases rapidly after injection of cholinesterase. Despite this, recovery from the neuromuscular block was slow due to slow elimination from the effect compartment.

To evaluate whether the concentration–effect relation differed from that of phenotypically normal patients, curve fitting of the data describing the concentration–effect model was performed and compared with data previously obtained in phenotypically normal patients. The pharmacokinetic–pharmacodynamic data from this latter group is derived from data presented in Østergaard *et al.*[16] Also, estimations of the concentrations of the combined active isomers correlating to 50% T_1 depression were performed (EC_{50}). Table 6 shows the fitted parameters in the Emax model for the combined active isomers. No significant differences in k_{eo} or $t_{1/2keo}$ were seen in phenotypically normal and phenotypically homozygous atypical patients for the combined effect of the two active isomers.

Discussion

Our main findings are that the clearances of mivacurium were significantly reduced and the elimination half-lives of the three isomers of mivacurium were prolonged in patients phenotypically homozygous for the atypical enzyme. This caused prolonged duration of action and recovery time of mivacurium. Injection of cholinesterase caused a normal or even high effective pChe activity, increased the clearance significantly, shortened the elimination half-lives, and enhanced recovery from neuromuscular block. However, in some patients, neuromuscular block recovered slowly. Injection of neostigmine increased the rate of recovery in the majority of patients. No difference in concentration–effect relation was found compared with phenotypically normal patients.

Enzyme Activity in Plasma before and after Injection of Cholinesterase

After administration of cholinesterase, pChe activity increased significantly in all patients, and in all but one, the effective pChe activity (= $pChe_{after} - pChe_{before}$) obtained was within the normal range used by the Danish Cholinesterase Research Unit.[11] The increase in pChe activity seen in our study is consistent with the findings of Naguib *et al.*[17]

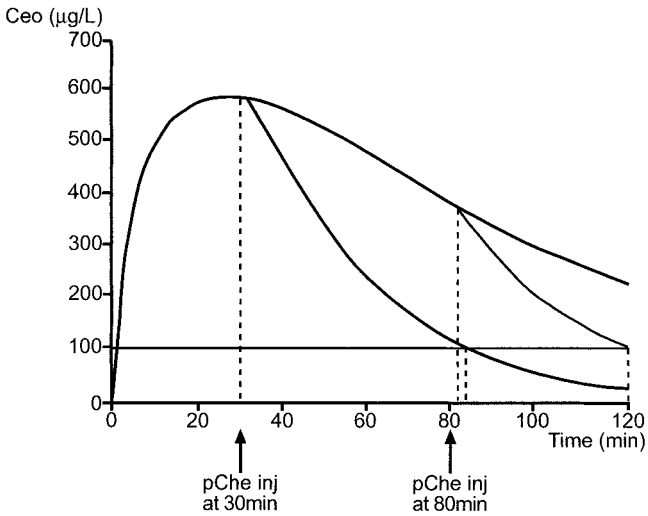

Fig. 2. Relation between the estimated effect compartment concentration of mivacurium (µg/l) and time (minutes) (*upper curve*) after injection of 0.15 mg/kg mivacurium. Also shown is the calculated effect compartment concentration following injection of cholinesterase after either 30 or 80 min (*two lower curves*). The minimum concentration for effect is 100 µg/l. The curves are simulated based on data from patients in the study. pChe = plasma cholinesterase.

Table 6. Equilibrium Rate Constant for Drug Removal from the Effect Site (k_{eo}), Half-life of Elimination from the Effect Compartment ($t_{1/2keo}$), Plasma Concentration at 50% T_1 Depression (EC_{50}), and Hill coefficient (γ) for Patients Homozygous for the Atypical pChe Gene

	k_{eo}, min^{-1}	$t_{1/2keo}$, min	EC_{50}, μg/l	γ
Homozygous patients	0.04 (0.02–0.16)	17.3 (4.3–34.7)	125 (23–214)	6.3 (2.1–17.5)
Normal patients	0.05 (0.02–0.10)	14.7 (6.7–39.0)	90 (30–140)	9.0 (3.0–29.6)

For comparison, values for phenotypically normal patients are given (unpublished data related to Gätke *et al.*[16]). Medians and ranges are given.
T_1 = time to reappearance of the first response to train-of-four stimulation.

Pharmacodynamics

Neuromuscular Blocking Effect of mivacurium before Injection of Cholinesterase. As expected, the duration of action of mivacurium was prolonged. T_1 had not reappeared in any of the patients at the time when cholinesterase was given 120 min after the administration of mivacurium. This is in accordance with spontaneous recovery data in a patient homozygous for the silent enzyme, in whom the time to recovery of T_1 was 274 min.[18]

Recovery of Neuromuscular Block after Injection of Cholinesterase. As expected, injection of cholinesterase increased the rate of recovery. However, no relation was found between the amount of injected cholinesterase and the rate of recovery of block, most probably because all patients obtained effective pChe activities above 550 U/l. Enzyme activities above this level do not seem to speed up the hydrolysis of either mivacurium or succinylcholine in phenotypically normal patients.[16,19] However, the number of patients included in the study was small, and that the three patients receiving the highest dose of cholinesterase received cholinesterase early after administration of mivacurium.

Judging from the pharmacokinetic data, a faster recovery from the late injection was to be expected (see Pharmacokinetics section). The time to T_1 10% was significantly shorter after the late injection of cholinesterase, but no significant difference was seen in time to reappearance of T_1, probably because of the small number of patients and the large variation in time to reappearance of T_1 (table 2).

There was a considerable delay from the injection of cholinesterase to the reappearance of T_1, especially after the early injection. This might be explained by the pharmacokinetics of mivacurium in those patients. When a mivacurium-induced block recovers spontaneously in phenotypically normal patients, mivacurium has been eliminated from plasma and tissues due to the rapid hydrolysis by pChe. In phenotypically homozygous atypical patients, the drug is not hydrolyzed in plasma but distributed to the different body compartments. At the time of the administration of cholinesterase, the amount of mivacurium in plasma represents represent only a small fraction of the total amount in the body. It takes some time before mivacurium, in other compartments including the neuromuscular endplate, is hydrolyzed—either after redistribution into plasma or by injected cholinesterase diffused out of plasma to the tissues. This might explain not only the initial slow recovery seen in this study after cholinesterase, but also that there is a difference in recovery time, depending on the time of injection of cholinesterase. This is illustrated in figure 2 with data derived from one of our patients. Thirty minutes after administration of mivacurium, the calculated concentration of mivacurium in the effect compartment (C_e) is significantly higher than 80 or 120 min later (upper curve). The effect of cholinesterase injected 30 and 80 min after mivacurium on C_e is illustrated (two lower curves). The time to minimal effective concentration (100 μg/l) is longer after an early injection of cholinesterase compared with a late injection.

Recovery of Neuromuscular Block after Injection of Neostigmine. Injection of neostigmine after administration of cholinesterase caused a fast recovery in the majority of patients with a shorter 25–75% interval. This decrease in 25–75% interval is in accordance with findings in phenotypically normal patients.[20] Several case reports have described unsuccessful administration of neostigmine at intense levels of block in phenotypically homozygous atypical patients,[5,7] whereas neostigmine seems to be effective after cholinesterase.[8,9] Our results document that neostigmine given after injection of cholinesterase at less intensive block is effective in increasing the rate of recovery. This is in accordance with the findings in phenotypically normal patients, in whom administration of neostigmine has different effects depending on the level of block. At intense block, neostigmine does not speed up recovery, a dual effect is seen when injected at 1–5% T_1, and recovery is increased when injected at greater than 10% T_1.[21–23]

Significance of Injected Cholinesterase for the Reaction to Supplementary Doses of Mivacurium. In the two patients receiving supplementary doses of mivacurium after administration of cholinesterase, recovery of T_1 was almost identical after the first and second doses of 0.03 mg/kg mivacurium, as well as after the 0.03 and 0.06 mg/kg doses. This is in accordance with the findings in phenotypically normal patients. However, the recovery time was longer, and the 25–75% interval was twice that seen in phenotypically normal patients.

Clinical Implications of the Pharmacodynamic Findings

As expected, spontaneous recovery from 0.15 mg/kg mivacurium is prolonged in patients phenotypically homozygous for the atypical variant, but recovery can be facilitated considerably by an injection of cholinesterase. The timing of the administration of cholinesterase is not crucial. Cholinesterase can be given both early and late after mivacurium. However, it often may take some time before the effect is seen, and the time to sufficient recovery (TOF ratio = 0.8) may be 30–60 min. Finally, in most patients with slow recovery of block after injection of cholinesterase, neostigmine speeds up recovery when injected at greater than 8% T_1. In some patients, however, the rate of recovery may still be slow.

Pharmacokinetics

Pharmacokinetics of Mivacurium before Injection of Cholinesterase. The clearances of the two active isomers (cis-trans and trans-trans) were lower than in phenotypically normal patients with normal or drug-induced low pCh activity[16] or low activity due to renal or hepatic failure,[24–26] whereas the clearance of the cis-cis isomer was less influenced.[2,17,24,25] Most probably, the clearance of the cis-cis isomer is predominantly renal, because a 50% reduction in clearance is seen in patients with end-stage renal failure.[24]

The elimination half-lives of the active isomers were significantly longer than in patients with normal pCh and also longer than in patients with drug-induced low pCh or disease.[16,24–26] Only data in patients receiving human cholinesterase late were used because valid data cannot be obtained with a sampling period of 30 min.

The initial volume of distribution in phenotypically homozygous patients was comparable to our findings in phenotypically normal patients using the same methodology and higher than that reported by Laurin *et al.*, who used arterial sampling.[16,27]

Pharmacokinetics of Mivacurium after Injection of Cholinesterase. Administration of cholinesterase caused an immediate decrease in plasma concentrations of all three isomers. After neostigmine, there was a slight increase in the concentration of the active isomers. This is consistent with previous findings and is most probably due the inhibiting effect of neostigmine on pCh and the diffusion of mivacurium back to plasma.[28]

The postcholinesterase clearance of the cis-trans isomer was higher than the clearance of the trans-trans isomer. This is in accordance with data from phenotypically normal patients.[16,24,25] This has also been observed *in vitro*.[29] The clearances of the cis-trans and trans-trans isomers were approximately 15 and 9 times higher, respectively, than before administration of cholinesterase and comparable to clearances in phenotypically normal patients previously studied using the same technique.[16] The clearances are slightly lower than after both short- and a long-lasting infusions of mivacurium in phenotypically normal patients.[3,24,25] The clearance of the cis-cis isomer increased 12-fold after injection of cholinesterase. This finding was unexpected because clearance of the cis-cis isomer is predominantly renal.[24] The resulting clearance is higher than reported earlier in phenotypically normal patients, where values from 3.8 to 5.4 $ml \cdot kg^{-1} \cdot min^{-1}$ have been found.[3,16,24,25]

The elimination rate constants of the active isomers increased 9- to 15-fold after administration of cholinesterase. The postcholinesterase values are comparable to reported data in phenotypically normal patients.[16] The elimination half-lives of the active isomers decreased, compared with the half-lives before administration of cholinesterase, whereas that of the cis-cis isomer decreased only 50%. The initial volume of distribution did not differ from parameters in normal patients.[16]

Pharmacokinetic–Pharmacodynamic Modeling

The estimated Emax, EC_{50}, k_{eo}, and γ (Hill coefficient) values did not differ from those found by us in phenotypically normal patients. The k_{eo} value is in accord with the findings of Schiere *et al.*[30] but higher than the EC_{50} and k_{eo} values reported by Laurin *et al.*[31] In the latter study and our study, a central compartment approach was used, but the methods used were not quite comparable. We cannot exclude that our estimations of these pharmacokinetic–pharmacodynamic parameters, at least in phenotypically normal patients, are flawed by the use of venous sampling as opposed to arterial, frequent sampling. In phenotypically normal patients, the hydrolysis of mivacurium in plasma is fast and starts immediately after injection. Measured plasma concentrations based on relatively sparse venous sampling may therefore not be representative of the changes in arterial concentrations, especially in the initial distribution phase.[32] In homozygous, atypical patients, however, hydrolysis is slow, and the plasma concentration remains high for a longer period of time. The data in these patients are therefore less flawed by the use of venous sampling. The major strength of this study is that the same approach was used in both groups of patients, indicating that if any differences are observed, they are the result of differences in the two groups of patients. Laurin *et al.*[31] have presented both a central approach and a peripheral link model, and the latter method seems to enable a more precise pharmacokinetic–pharmacodynamic modeling. Also, Schiere *et al.*[30] has presented a model with an interstitial space compartment interposed between plasma and the effect compartment. However, we decided to use the conventional method to make comparisons with data in phenotypically normal patients. All in all, we think that the pharmacokinetic–pharmacodynamic analyses support our pharmacokinetic findings and our conclusion that the longer duration of action of mivacurium in patients phenotypically homozygous for

the atypical enzyme is due to pharmacokinetic changes caused by a less efficient pCh enzyme.

In conclusion, in patients phenotypically homozygous for the atypical variant, clearance of mivacurium is reduced and the elimination half-lives are prolonged, causing a markedly prolonged duration of action of mivacurium. Injection of human cholinesterase increases pCh activity, leading to a 9- to 15-fold increased clearance and a shorter elimination half-life of mivacurium. This in turn shortens the duration of action of mivacurium three to four times. Although the plasma concentration decreases rapidly, there is a delay from the time of injection of cholinesterase to the reappearance of T_1, most pronounced after early injection of cholinesterase. Neostigmine administered after cholinesterase increases the rate of recovery further, although recovery may be slow in some patients. The elimination rate constant from the effect compartment (k_{eo}) seems to be the same in patients phenotypically homozygous for the atypical variant and phenotypically normal patients.

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