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NMDA 和非 NMDA 受体介导猫脊髓伤害性和非伤害性信息传递

R 965.2

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摘要 用多管微电极离子电泳技术研究猫脊髓背角神经元活动. 80%的神经元被兴奋性氨基酸 NMDA、使君子氨酸、卡因酸或 *DL*-高磺丙氨酸（DLH）激活. NMDA 受体拮抗剂 APV 和氯胺酮，非 NMDA 受体拮抗剂 DNQX 以及广谱拮抗剂 4-羟基喹啉酸(Kyn)均抑制神经元的伤害性反应. Kyn 抑制非伤害性反应大大强于氯胺酮. NMDA 和非 NMDA 受体均参与介导脊髓伤害性反应，非伤害性信息主要由非 NMDA 受体介导.

关键词 伤害性感受器；脊髓；氨基酸受体；*N*-甲基-*D*-门冬氨酸；使君子氨酸；卡因酸；*DL*-高磺丙氨酸

高磺丙氨酸

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Midazolam pharmacokinetics and electroencephalographic changes in eight Chinese men[1]

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ABSTRACT Eight Chinese healthy male volunteers aged 27±*s* 4 a were injected iv midazolam (Mid) 15 mg. Blood samples were collected at 0, 2, 5, 7, 10, 20, 30, 45, 60, 90, 120, 180, and 240 min. A HPLC method was established for determining the Mid concentrations in serum. The concentration-time data was fitted with biexponential curve. Pharmacokinetic parameters were: $T_{\frac{1}{2}\alpha}$=6.8±2.5 min, $T_{\frac{1}{2}\beta}$=118±27 min, V_c=25±7 L, Cl=393±79 ml·min^{-1}, V_{ss}=59±13 L, $AUC_{0-\infty}$=39.6±8.6 g·min·L^{-1}. The electroencephalogram (EEG) showed a decrease in α activity and an increase in β activity. The EEG pattern reverted toward baseline after 2—3 h.

Pharmacokinetic and EEG findings suggest that Mid is a preferable anesthesia inducing agent.

KEY WORDS midazolam; pharmacokinetics; high pressure liquid chromatography; electroencephalography

Midazolam (Mid) is used in anesthesia for premedication, induction of anesthesia, and intraoperative hypnosis[1,2]. But there has

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been no report on Mid kinetics in Chinese. Since evaluation of the pharmacokinetic properties of Mid is important for its clinical use, a sensitive HPLC method was established for quantitation of Mid. Pharmacokinetic course of Mid following a single iv dose was studied in 8 Chinese volunteers and EEG changes were observed simultaneously.

MATERIALS AND METHODS

Volunteers After personal approval and written consent were obtained, 8 healthy men aged 27±s 4 a, weighing 61 ± s 4.5 kg, participated in the study. None was receiving benzodiazepines; all were refrained from alcohol, caffeine-containing beverages and food from the midday prior to the investigation.

Study design All studies were undertaken at 4:00 pm in a same room. The subject lay on a bed. Two iv catheters were inserted: One for blood sampling and the other in another arm for injection of Mid. Mid maleate (Roche) 15 mg dissolved in 20 ml of 0.9% saline was injected in 2 min. Wakening was defined as eyes opened and consciousness resumed

Blood sampling Venous blood samples (3 ml) were taken at 0, 2, 5, 7, 10, 15, 20, 30, 45, 60, 90, 120, 180, and 240 min after infusion was finished. The serum was stored at −40℃.

Drug effect measurement For recording EEG, 5 AgCl electrodes were fixed to scalp. Electrode positions were frontal pole (Fp1, Fp2), occipital (O1, O2) and central zero (C_z), according to the international 10−20 system. The normal EEG waves and heart beats were recorded on paper 5 min before iv Mid. The EEG and heart beats were simultaneously recorded 10 min after iv and then recored 1 min every 10 min afterwards with a 17-channel EEG instrument (Nihon Kohden 4217) and EEG signals were also recorded in SUN-386 computer for subsequent off-line analysis. The injection time, time of onset of sleep, time of wakening, and respiratory rate were calculated.

Assay of Mid Mid concentrations in serum were measured by reverse HPLC[3,4]. Serum aliquots of 0.5 ml, to which diazepam was added as internal standard, were mixed with 25 μl NaOH 1 mol · L^{-1} in a contrifuge tube. The sample was extracted with diethylether 8 ml for 1 min with a whirl-mixer and then centrifuged at 2000×g for 5 min. Organic phase (5 ml) was evaporated to dryness. The residue was dissolved in methanol 100 μl and 20 μl was injected into a modular Shimadzu HPLC system, consisting of LC-6A pump, SPD-6AV uv detector, C-R6A integrator, and a stainless-steel column (250 mm×2 mm) prepacked with ultrasphere TM C18, 3 μm. The uv absorbance detection was set at 220 nm. The mobile phase consisted of methanol-acetonitrile-water (35 : 35 : 30, vol : vol) to which n-butylamine 1.2 ml per liter of mobile phase was added. Final pH of the mobile phase was adjusted to 7.0 with HAc and the flow rate was 1 ml · min^{-1}. The retention times of diazepam and Mid were 6.5 and 8.3 min, respectively. The detection limit was approximately 10 ng Mid/ml serum. The CV in this study was <5%.

Data analysis The conentration-time data were fitted using weighted ($1/c^2$) least-squares nonlinear regression analysis with the software package PKBP-N1. The pharmacokinetic parameters were calculated for each volunteer using standard equations. EEG tracings before and after iv were compared.

RESULTS

The 8 subjects fell asleep at 58±13 s. The time of wakening ranged 106−158 min. The Mid concentration at the time of wakening was 91±14 ng · ml^{-1}.

Heart rate and respiratory rate did not change during or after iv Mid. In all subjects, disappearance of Mid from serum was consistent with the two exponential curve (Fig 1 and Tab 1). The initial distribution half-life of Mid was rapid, with $T_{\frac{1}{2}\alpha}$ of 6.8±2.6 min. The elimination half life ($T_{\frac{1}{2}\beta}$) was 118 ±27 min. Mid distribution was reasonably extensive, with $V_{d_{ss}}$ values averaging 59±13 L. Mean total clearance was 393 ml · min^{-1}.

The EEG of 8 men were similar. After 60−90 s from the start of 2 min injection, β activity (above 13 Hz) emerged with a decrease of α activity. The β activity increased rapidly to a maximum 2−5 min from the end of iv, with the disappearance of α rhythm (8−

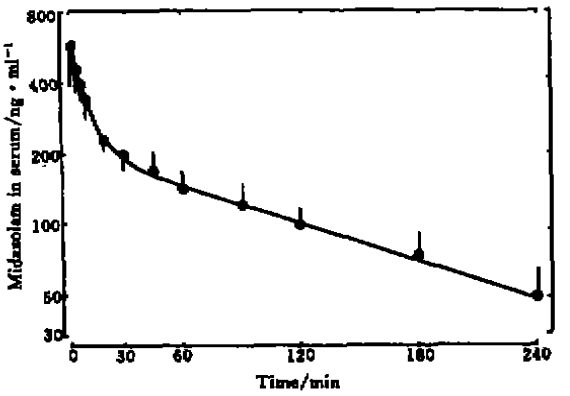

Fig 1. Midazolam concentration in serum of 8 men after iv midazolam 15 mg. $\bar{x}\pm s$.

13 Hz). Thereafter, β activity declined rapidly. The EEG pattern mainly consisted of β activity with well-modulated spindling. Then the frequency and height of EEG waves decreased gradually. Prior to wakening, some measurable α rhythm reappeared. The men waked after 2—3 h and EEG pattern reverted to the baseline (Fig 2).

DISCUSSION

The results showed that Mid had only a minor effect on the respiratory and cardiovascular system. All men waked up in 106—158 min which was much longer than the duration reported by Breimer *et al*[4]. In a study similar to ours Breimer *et al* gave 15 mg Mid as a 5-min infusion to 8 young male volunteers weighing 75±4 kg, they found that all volunteers waked up in 60—70 min. The differences of the results might be explained by which method was used. The studies of Breimer *et al* took place in the morning and vigilance was maintained by regular verbal contact. Our study took place in the afternoon and no vigilance was maintained, so the subjects slept longer easily.

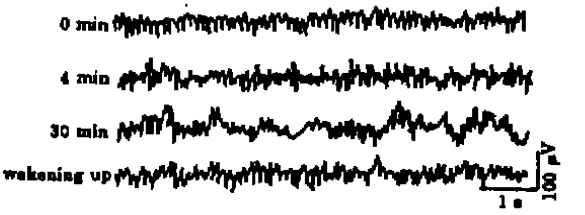

Fig 2. EEG tracing (Fp 1) from a man after iv midazolam 15 mg.

Kinetic findings showed that Mid was a short-acting benzodiazepine derivative that was rapidly cleared from plasma. After a quick distribution of α phase ($T_{\frac{1}{2}\alpha}=6.8$ min), elimination of Mid proceeded with a half life averaging 119 min in Chinese men. This was consistent with that reported in similar studies[1,2,4].

Tab 1. Pharmacokinetic parameters for midazolam in individual subjects.

	α min^{-1}	β min^{-1}	$T_{\frac{1}{2}\alpha}$ min	$T_{\frac{1}{2}\beta}$ min	K_{21} min^{-1}	K_{12} min^{-1}	K_{10} min^{-1}	V_c L	V_{dss} L	Cl L · min^{-1}	AUC g · min · L^{-1}
1	0.079	0.0049	8.8	142	0.033	0.039	0.012	32	70	0.37	40.1
2	0.082	0.0045	8.4	153	0.038	0.039	0.099	31	63	0.30	49.5
3	0.244	0.0049	2.8	141	0.044	0.178	0.027	11	52	0.27	55.2
4	0.138	0.0066	5.0	104	0.048	0.078	0.019	24	55	0.44	34.5
5	0.063	0.0053	11.0	130	0.022	0.031	0.015	33	81	0.52	28.7
6	0.122	0.0097	5.7	71	0.067	0.047	0.018	24	38	0.41	36.8
7	0.099	0.0069	7.0	100	0.041	0.048	0.017	25	53	0.41	36.7
8	0.120	0.0065	5.8	106	0.040	0.066	0.020	22	61	0.42	35.4
$\bar{x}$	0.118	0.0062	6.8	118	0.041	0.066	0.028	25	59	0.39	39.6
s	0.057	0.0017	2.6	27	0.013	0.048	0.029	7	13	0.08	8.6

Clearance was 393 ml · min^{-1} in Chinese men which was similar to what Breimer *et al* reported (Cl = 391 ml · min^{-1}). A previous study[6] in intensive care patients showed that continuous intravenous infusion had no effect on the value of clearance, which was 415 ml · min^{-1} and similar to the results in healthy volunteers Heizmann *et al* reported[1]. So the clearance of Mid did not change easily and there was no difference between Chinese young men and European yound men.

Mid tissue distribution was smaller in Chinese men than that in European young men[4]. In a study for investigation of the influnce of old age and obesity on the kinetics of Mid, Greenblatt *et al* found that the volume in young male volunteers (weighing 69±1.7 kg) was 92 L, and the volume in obesity volunteers was much larger. Furthermore, in intensive care patients received continuous intravenous infusion[6], the volume of Mid increased three times. So the volume of Mid can change with various conditions and there was difference between Chinese men and Europeans.

Sudden administration of benzodiazepines to humans produced a shift in the EEG pattern, usually characterized by increased activity in the β frequency region, ranging from 13 to 30 Hz[5]. The study considered β activity as the qualitative assessment of central effects of Mid. We found that Mid produced EEG effects of rapid onset. Mid effects were maximal at 2—5 min after the injection. EEG pattern reverted rapidly toward baseline in 2—3 h, *ie*, EEG effects became indistinguishable from baseline before plasma concentratins fell to zero. The results were similar to those reported [5] Rapid onset and offset properties may be an important reason to consider Mid to be an anesthestic induction agent preferable to other benzodizepines.

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8 位健康男性中咪唑安定的药物动力学及脑电图变化

R971.2

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摘要 8 位健康男性，iv 咪唑安定 15 mg 后每隔一定时间抽取血样，咪唑安定浓度用 HPLC 法测定，血清药物浓度—时间数据用双指数曲线拟合，主要动力学参数如下：$T_{\frac{1}{2}\alpha}$=6.8±2.6 min，$T_{\frac{1}{2}\beta}$=118±27 min，V_c=25±7 L，Cl=393±79 ml · min^{-1}，V_{dss}=59±13 L，$AUC_{0-\infty}$=39.6±8.6 mg · min · L^{-1}。药后脑电图(EEG)的变化主要为 α 波减少，β 波明显增多。药物动力学和 EEG 分析表明咪唑安定是理想的麻醉诱导剂。

关键词 咪唑安定；药物动力学；高压液相色谱法；脑电图