

Pharmacokinetics of theophylline metabolites in 8 Chinese patients¹

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KEY WORDS theophylline; aminophylline; xanthines; pharmacokinetics

AIM: To study theophylline metabolites pharmacokinetics in patients after a therapeutic dose. **METHODS:** Eight adult patients with mild bronchial asthma and normal liver function were infused aminophylline intravenously ($6.6 \mu\text{mol} \cdot \text{kg}^{-1}$). The plasma concentrations of theophylline and its 4 metabolites: 1,3-dimethyluric acid (DMUA), 3-methylxanthine (3-MX), 1-methyluric acid (MUA), and the intermediate 1-methylxanthine (1-MX) were monitored by HPLC throughout 24 h. **RESULTS:** The plasma concentration of DMUA was the highest one among the 4 metabolites. 3-MX showed the slowest elimination rate. The plasma concentration of 1-MX throughout a 24-h period showed that there was a picking up of 1-MX (from $0.04 \mu\text{mol} \cdot \text{L}^{-1}$ to $1.05 \mu\text{mol} \cdot \text{L}^{-1}$) in the next morning. **CONCLUSION:** The formation of DMUA was the main metabolites. During night there was an accumulation of 1-MX.

Theophylline undergoes extensive metabolism in liver. More than 80 % of the dose appears in urine as 1, 3-dimethyluric acid (DMUA), 3-methylxanthine (3-MX), 1-methyluric acid (MUA)^[1]. 1-MX is a metabolic intermediate in the formation of MUA. The elimination of theophylline was slow at night. These differences have yet to be explained in terms of changes in specific metabolic pathways. 1-MX, an intermediate metabolite of theophylline, have never been reported in plasma throughout 24 h. Little can be concluded regarding the relationship between elimination kinetics of theophylline metabolites and theophylline pharmacokinetics. Our studies were designed to reveal the elimination kinetics

relationship between theophylline and its metabolites under a therapeutic dose in adult patients.

MATERIALS AND METHODS

Reagents Aminophylline (Hangzhou Minsheng Pharmaceutical Co, China, containing 80 % theophylline and 15 % ethylenediamine, lot No 931202). Standards of theophylline and its 4 metabolites were given from Professor Donald J BIRKETT, Department of Clinical Pharmacology, Flinders University of South Australia and Flinders Medical Centre, South Australia. Other reagents were of HPLC grade.

Chromatography A Beckman 344 HPLC system was used, the column (Spherisorb C₁₈, 5 μm , 4.6 mm \times 250 mm) was purchased from Beckman Co.

The plasma concentrations of theophylline and its 4 metabolites were assayed by HPLC^[4]. The plasma samples were filtered through a pre-column packed by ourselves and then evaporated to dryness under a stream of nitrogen at 40 °C, the residue reconstituted in 0.2 mL of mobile phase. The mobile phase consisted of methanol, acetonitrile, and sodium acetate (9:4:30).

Subjects Eight patients (aged 22 - 62 a, $40 \pm s 14$) with mild bronchial asthma and normal liver functions were treated with aminophylline. None were taking chronic medication. The patients were asked to abstain from coffee and tea for 72 h before medication.

Study design Patients were infused aminophylline intravenously ($6.6 \mu\text{mol} \cdot \text{kg}^{-1}$) from 8:00 to 8:30. Blood samples were drawn at 0, 1, 2, 4, 12, 24 h after administration. Plasma samples were stored at -40 °C and analyzed within 1 wk.

RESULTS

The chromatograms of theophylline and 4 metabolites from patient plasma samples were shown in Fig 1. Theophylline and 4 metabolites were separated well.

The regression equations and r values for

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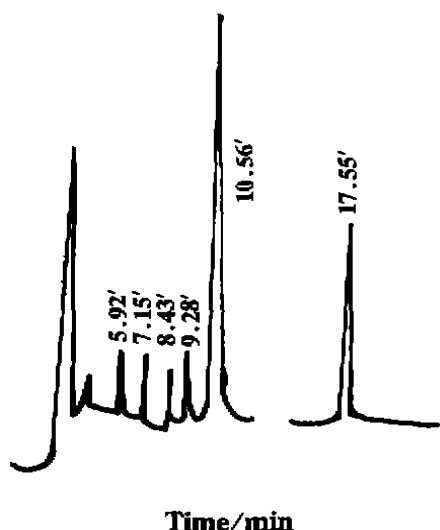


Fig 1. Chromatogram of theophylline (17.55 min) and its metabolites: DMUA (9.28 min), 1-MX (8.43 min), 3-MX (7.15 min), MUA (5.92 min). Attenuation = 2^o before 12 min, and 2^o after 12 min.

standard curves followed by Tab 1. All the *r* values ≥ 0.99 . The lower detection limit was 63.07 nmol. Recovery rates for the different metabolites ranged from 74.7 % to 99.1% (Tab 2). Inter- and intra-day variability of the assay for all compounds was less than 10 %. Drugs commonly coadministered with theophylline did not interfere with the assay.

Tab 1. Mean regression equations and *r* values for standard curves.

Compound	\bar{y}	<i>r</i>
MUA	0.4325 X^* + 0.0433	0.9977
3-MX	0.5988 X^* - 0.0106	0.9985
1-MX	0.6568 X^* - 0.0100	0.9993
DMUA	0.6492 X^* - 0.0027	0.9998
Theophylline	0.9983 X^* - 0.1200	0.9998

X^* : peak height of plasma concentrations

The profiles based on the mean plasma concentration-time data from 8 patients were shown in Fig 2. DMUA had the highest concentration among 4 metabolites. The clearance of 1-MX was very fast within 12 h and the concentration of 1-MX came to its nadir at 20:00. But at 24 h (8:00 next morning), it picked up to a higher point. All the others had no such changes.

Tab 2. Recovery and sensitivity limits of assay for theophylline and its metabolites in plasma *n* = 15. $\bar{x} \pm s$.

Compound	Recovery/%	Sensitivity limit/nmol·L ⁻¹
Theophylline	98 ± 7	126.1
DMUA	97 ± 8	63.1
1-MX	87 ± 8	63.1
3-MX	99 ± 5	63.1
MUA	75 ± 9	63.1

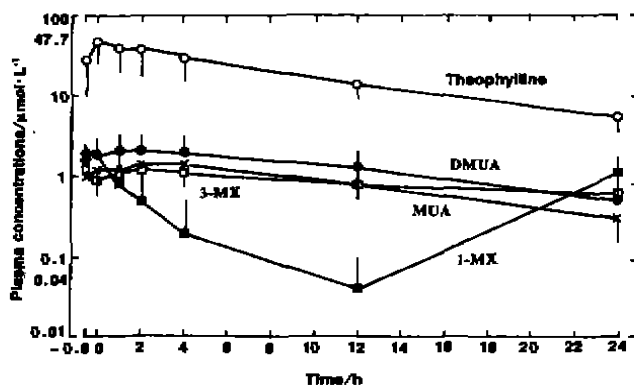


Fig 2. Plasma concentrations of theophylline and 4 metabolites (*n* = 8 patients).

The elimination rates of theophylline and its metabolites were estimated by regression analysis based on the linear relationship between log-concentration vs time at the terminal (after 4 h) of the profiles. The elimination of theophylline and DMUA and MUA were similar, 3-MX had the slowest elimination rate (Tab 3).

Tab 3. Elimination rate of theophylline and 4 metabolites estimated from the mean plasma concentrations by linear regression at the end of the profile.

Parameters	Theophylline	DMUA	3-MX	1-MU	1-MX
K/h^{-1}	0.084	0.066	0.036	0.081	0.298
$t_{1/2}/h$	8.25	10.45	19.28	8.52	2.32

DISCUSSION

The assay of theophylline and 4 metabolites in plasma for our study was highly reproducible and specific. It would be very important in the further study of the metabolism of theophylline.

The plasma concentration of DMUA was the highest one among the 4 metabolites before 12 h

after a routine dose. This suggested that DMUA was the main metabolite of theophylline. 3-MX had the slowest elimination rate, it supported that the formation rate of 3-MX was capacity limited^[5].

1-MX is an intermediate between theophylline and MUA. The rate of metabolism of 1-MX to MUA greatly exceeded the rate at which it was formed^[2]. The concentration of 1-MX in plasma was very low. We reported plasma concentration of 1-MX throughout a 24-h period and found a picking up of 1-MX concentration at 24 h after a routine dose. There were 2 reasons: the converting of 1-MX to MUA decreased and/or theophylline to 1-MX increased. However, no matter what reasons, 1-MX accumulated during night, which might be one of the reasons of that theophylline clearance declined at night.

The metabolites 5-acetylamino-6-formylamino-3-methyluracil (AFMU)/(1-MX + MUA) ratio of caffeine was used to characterize the *N*-acetyltransferase phenotype^[6]. The results varied within independent studies that concerned on the same race^[6]. There were no clear explains. Theophylline and caffeine have a similar construction and a same biotransformation of 1-MX to MUA. In our study, 1-MX had a great change of concentrations throughout 24 h, which led to a vary ratio of AFMU/(1-MX + MUA), and at last the results were different. It was necessary to pay attention to the circadian metabolism before using a drug as a probe to reveal the activities of enzymes.

REFERENCES

- 1 Zhang ZY, Fasco MJ, Kamirsky LS.
Determination of theophylline and its metabolites in rat liver

microsomes and human urine by capillary electrophoresis.
J Chromatogr Biomed Appl 1995; 655: 201-8.

- 2 Birkett DJ, Mivers JO, Attwood J.
Secondary metabolism of theophylline biotransformation products in man route of formation of 1-methyluric acid.
Br J Clin Pharmacol 1983; 15: 117-9.
- 3 Lin L, Lu W, Zhang Q, Chen WD, Chen LY.
Circadian rhythms of plasma level on controlled release aminophylline tablet in normal subjects.
Chin J Hosp Pharm 1989; 9: 385-7.
- 4 Rasmussen BB, Nielsen KK, Brosen K.
Determination of theophylline metabolites in human liver microsomes by high-performance liquid chromatography.
Anal Biochem 1994; 222: 9-13.
- 5 Tang-Liu DS, Williams RL, Riegelman S.
Nonlinear theophylline elimination.
Clin Pharmacol Ther 1982; 31: 358-69.
- 6 Kalow W, Tang BK.
The use of caffeine for enzyme assays: a critical appraisal.
Clin Pharmacol Ther 1993; 53: 503-14.

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8例中国病人茶碱代谢物的药物动力学¹

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关键词 茶碱; 氨茶碱; 黄嘌呤类; 药物动力学

目的: 研究病人常规剂量茶碱治疗后代谢物的药理学。 **方法:** 病人静滴茶碱 ($6.6 \mu\text{mol} \cdot \text{kg}^{-1}$)。 HPLC 法测定给药前后 24 h 茶碱及其代谢物: 1,3-二甲基尿酸 (DMUA), 3-甲基黄嘌呤 (3-MX), 1-甲基尿酸 (MUA), 中间代谢产物 1-甲基黄嘌呤 (1-MX) 的浓度。 **结果:** DMUA 是代谢物中浓度最高的。 3-MX 的清除速率最低。 1-MX 很快转化成 MUA, 体内浓度很低, 但是, 翌晨, 1-MX 又回升到一个较高的浓度 (从 $0.04 \mu\text{mol} \cdot \text{L}^{-1}$ 上升到 $1.05 \mu\text{mol} \cdot \text{L}^{-1}$)。 **结论:** DMUA 是茶碱的主要代谢产物; 在夜间 1-MX 浓度积蓄, 这是茶碱在夜间消除率下降的原因之一。

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