Pharmacokinetics of a New Thromboxane A₂ Receptor Antagonist, S-1452, and Its Effect on Platelet Aggregation in the Elderly

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The effect of age on the pharmacokinetics of a new thromboxane A₂ (TXA₂) receptor antagonist, S-1452, and the inhibitory effect on platelet aggregation were examined in young (24~31 years, n=8) and elderly (68~78 years, n=8) male subjects. Ten mg of S-1452 or placebo was given orally in a single-blind cross-over design.

Hepatic function estimated by indocyanine green (ICG) test decreased in the elderly [elimination rate constant of ICG; elderly: 0.161 (mean) vs. young: 0.192/min, $p < 0.05$]. The maximum plasma concentration of S-1452 tended to be greater in the elderly than in the young [elderly: 28.7 (mean), young: 16.4 ng/ml]. The ratio of the metabolites to unchanged compound in the C_{max} was significantly lower in the elderly than in the young [elderly: 1.1 (mean) vs. young: 1.7, $p < 0.05$]. The inhibitory effect of S-1452 on platelet aggregation *ex vivo* induced by U-46619, a TXA₂ receptor agonist, persisted up to 6 and 9 hours after dosing in the young and elderly subjects, respectively.

These results suggest that the plasma concentration of S-1452 is higher, probably due to the reduced conversion of S-1452 to the metabolites, and the inhibition on platelet aggregation is prolonged in the elderly.

Key words: S-1452, thromboxane A₂ receptor antagonist, elderly

Introduction

The pharmacokinetics of many drugs are altered in the elderly. These alterations are primarily due to the decreases in hepatic metabolism or renal elimination, or both, which occur with age¹⁾. Consequently, drug responses are enhanced and adverse reactions are often ob-

served in these subjects¹⁾. Therefore, it is important that the effect of age on pharmacokinetics and pharmacodynamics of a drug be studied in the elderly.

S-1452, calcium 5 (Z)-1R, 2S, 3S, 4S, 7-[3-phenylsulfonyl-amiobicyclo[2. 2. 1]hept-2-yl]-5-heptenoate hydrate, is a highly specific thromboxane A₂ (TXA₂) receptor antagonist²⁾, which might be used for the treatment of TXA₂-related diseases such as myocardial ischemia and bronchial hyperresponsiveness^{3,4)}. Previous

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Table 1 Profiles of the Young and Elderly

Subject	Age (years)	Body weight (kg)	ICG-Kel (min ⁻¹)	Clcr (ml·min ⁻¹)	S-albumin (mg/dl)
Young (n=8)	26 (22~31)	67 (51~82)	0.192 (0.157~0.227)	96 (61~132)	4.15 (4.00~4.30)
Elderly (n=8)	71** (65~77)	63 (49~78)	0.161* (0.114~0.208)	72* (46~98)	3.84** (3.46~4.22)
Normal range			0.168~0.206	62~108	

* : $p < 0.05$, ** : $p < 0.01$ compared with the young

Mean (and 95% confidence intervals)

ICG-Kel : elimination rate constant of indocyanine green

Clcr : creatinine clearance

studies have shown that S-1452 is rapidly absorbed and extensively metabolized in young subjects^{5,6)}.

In the present study, S-1452 was given orally to young and elderly subjects. The pharmacokinetics of S-1452 and the inhibitory effects on platelet aggregation *ex vivo* in the elderly were compared with those in the young.

Material and Methods

Sixteen male subjects were studied (Table 1). Serum albumin in the elderly was significantly lower than that in the young. None of the young had a history of cardiovascular, renal or hepatic disease. Physical examination, electrocardiogram (ECG) and laboratory (biochemical, hematological and urological) screening examinations were normal before inclusion. Four elderly subjects had one or more of the following mild abnormalities : one subject ; hypertension (166/94 mmHg), one subject ; ECG abnormality (CRBBB), two subjects ; hypertriglyceridemia (190, 231 : normal range 40~185 mg/dl), one subject ; hyperglycemia (FBS 140 : normal range 70~120 mg/dl) and one subject ; renal dysfunction (S-Cr 1.3 : normal range 0.6~1.2 mg/dl). The other four elderly subjects did not have any abnormal finding. The subjects gave informed written

consent to the study's purpose and methods. The study was approved by the Review Board of Shionogi Institute for Clinical Pharmacology.

The subjects were admitted to a special hospital ward, and were confined to the study area 12 hours before and during the study. Alcoholic beverages, coffee, tea and smoking were not allowed on the day before and during the study. Subjects did not take any cyclooxygenase inhibitor, such as aspirin or indomethacin, for 10 days before the study and until the end of the trial.

A 10 mg tablet of S-1452 and also placebo (Shionogi & CO., Ltd., Osaka, Japan) were used in this study. After overnight fasting, S-1452 (10 mg) or placebo was given orally with 100 ml water at 9 : 00 am by a single-blind design. Subjects were randomly divided into two groups. Group A (young : n=4, elderly : n=4) received placebo on Day 1 and S-1452 on Day 8 while Group B (young : n=4, elderly : n=4) received S-1452 on Day 1 and placebo on Day 8. Subjects remained in supine position for 3 hours and did not take any food for 4 hours after dosing. Physical examinations were done, and blood pressure and ECG were monitored at frequent intervals. The subjects were questioned about adverse symptoms. Laboratory

examinations for blood, urine and stool were done before and at the end of the study. Furthermore, these examinations were also done at some times during 3 weeks after the final dose on day 8.

Blood samples for assay of plasma S-1452 and its main metabolites (inactive), bisnor-(+)-S-145 and tetranor-(+)-S-145, were obtained before and at 0.5, 1, 2, 3, 4, 6, 9, and 24 hours after dosing. Platelet aggregation *ex vivo* was determined just before and at 1, 3, 6, 9 and 24 hours after treatment. Urine samples for analysis of S-1452 and the metabolites were collected between 0 and 4, 4 and 12, and 12 and 24 hours after dosing.

Plasma and urinary concentrations of S-1452 (unchanged and glucuronide), bisnor-(+)-S-145 and tetranor-(+)-S-145 were measured by capillary gas chromatography^{5,6)}. Platelet aggregation using platelet rich plasma (PRP) was determined by a method described previously⁷⁾. For the aggregating agent, 1 μ M U-46619, a TXA₂ receptor agonist (Shionogi & Co., Ltd., Osaka, Japan) was used. Maximum platelet aggregation in response to 1 μ M U-46619 was more than 50% in each subject when these volunteers were selected.

To determine the elimination rate constant of indocyanine green (ICG-Kel) in each subject, plasma samples were obtained at 5, 10 and 15 minutes after the rapid intravenous injection of 0.5 mg/kg, and were analyzed by the method of Caesar et al⁸⁾. Creatinine clearance (Clcr) was determined using the 24-hour urine sample during placebo treatment.

The maximum plasma concentration ($C_{\max}$) and time to maximum concentration ($t_{\max}$) were calculated directly from the plasma concentration data for each subject. The area under the plasma concentration-time curve from 0 to 24 hours (AUC_{0-24}) was determined

by the linear trapezoidal rule. The terminal elimination rate constant (Kel) was obtained by curve fitting of the plasma concentrations to a one-compartment open model. The elimination half-life ($t_{1/2}$) was calculated as: $t_{1/2} = \ln 2 / \text{Kel}$.

Data are expressed as the mean and 95% confidence intervals. Statistical analysis was performed by paired t-test.

Results

The values of ICG-Kel and Clcr in the elderly were significantly smaller than those in the young (Table 1).

S-1452 was rapidly absorbed after oral dosing and its plasma peak was observed at 0.7 and 0.5 hour in the young and elderly, respectively (Fig. 1 and Table 2). Thereafter, the drug was rapidly decreased and was not detected at 6 hours in the plasma. The $C_{\max}$ and AUC_{0-24} of S-1452(unchanged) in the elderly were greater than those in the young, but the difference did not reach statistical significance. The $t_{1/2}$ in the elderly was similar to that in the young. Mean plasma concentrations of total S-1452 (unchanged + glucuronide) and inactive metabolites, bisnor-(+)-S-145 and tetranor-(+)-S-145, are shown in Figure 2. The $C_{\max}$ and AUC_{0-24} of total S-1452 were also significantly greater or tended to be greater in the elderly than in the young (Table 2). There was no significant difference in any pharmacokinetic parameter of the metabolites between the young and elderly. The mean ($\pm$ SD) AUC_{0-24} of S-1452 (total) + the metabolites of the young and elderly did not differ significantly [young: 270 ($\pm$ 58), elderly: 311 ($\pm$ 114) ng $\cdot$ hr/ml]. No significant difference was observed in the cumulative amount of urinary S-1452 and the metabolites in the 4-, 12-, or 24-hour urine sample (Fig. 3).

Fig. 1 Plasma concentrations of S-1452 (unchanged) after administration of 10 mg in the young ($n=8$, $\bigcirc$ — $\bigcirc$) and elderly ($n=8$, $\bullet$ — $\bullet$) Mean $\pm$ SD
: $0.05 < p < 0.10$ compared with the young

Table 2 Pharmacokinetic Parameters of S-1452 and Its Metabolites after Administration of 10 mg in the Young and Elderly

Compound	Parameter	Subject	
		Young ($n=8$)	Elderly ($n=8$)
S-1452 (unchanged)	$C_{\max}$ ($\text{ng}\cdot\text{ml}^{-1}$)	16.4 ($-1.5 \sim 34.3$)	28.7 ($-10.0 \sim 67.3$)
	$t_{\max}$ (hr)	0.7 ($0.2 \sim 1.2$)	0.5
	$t_{1/2}$ (hr)	0.5 ($0 \sim 1.0$)	0.5 ($0.3 \sim 0.7$)
	AUC_{0-24} ($\text{ng}\cdot\text{hr}\cdot\text{ml}^{-1}$)	21.2 ($3.2 \sim 39.1$)	30.2 ($-0.6 \sim 61.0$)
S-1452 (total)	$C_{\max}$ ($\text{ng}\cdot\text{ml}^{-1}$)	37.2 ($23.4 \sim 51.0$)	78.7* ($-14.0 \sim 171.4$)
	$t_{\max}$ (hr)	0.7 ($0.2 \sim 1.2$)	0.5
	AUC_{0-24} ($\text{ng}\cdot\text{hr}\cdot\text{ml}^{-1}$)	52.8 ($31.4 \sim 74.3$)	86.4# ($1.8 \sim 171.0$)
Bisnor-(+)-S-145	$C_{\max}$ ($\text{ng}\cdot\text{ml}^{-1}$)	41.9 ($27.7 \sim 56.1$)	46.0 ($4.6 \sim 87.4$)
	$t_{\max}$ (hr)	1.0	1.0
	AUC_{0-24} ($\text{ng}\cdot\text{hr}\cdot\text{ml}^{-1}$)	182.8 ($105.3 \sim 206.4$)	166.3 ($47.1 \sim 285.4$)
Tetranor-(+)-S-145	$C_{\max}$ ($\text{ng}\cdot\text{ml}^{-1}$)	18.3 ($9.9 \sim 26.6$)	28.4 ($-10.7 \sim 67.5$)
	$t_{\max}$ (hr)	0.8 ($0.2 \sim 1.3$)	0.6 ($0.2 \sim 0.9$)
	AUC_{0-24} ($\text{ng}\cdot\text{hr}\cdot\text{ml}^{-1}$)	34.6 ($13.5 \sim 55.8$)	57.9 ($-14.8 \sim 130.5$)

Mean (and 95% confidence intervals)

: $0.05 < p < 0.10$, * : $p < 0.05$ compared with the young

$C_{\max}$: plasma maximum concentration, $t_{\max}$: time to maximum concentration,

$t_{1/2}$: elimination half-life, AUC_{0-24} : area under the plasma concentration-time curve from 0 to 24 hours

Fig. 2 Plasma concentrations of S-1452 (total) and its metabolites, bisnor-(+)-S-145 (M_1) and tetranor-(+)-S-145 (M_2), after administration of 10 mg in the young ($n=8$, $\bigcirc$ — $\bigcirc$) and elderly ($n=8$, $\bullet$ — $\bullet$) Mean $\pm$ SD
* $p < 0.05$ compared with the young

The ratio of the metabolites (β -oxidation and hydroxylation) to unchanged compound in the $C_{\max}$ and AUC_{0-24} was significantly smaller or tended to be smaller in the elderly than in the young (Fig. 4). There was no significant difference in the urinary excretion of S-1452 or the metabolites between the young and elderly.

The inhibitory effect of S-1452 on platelet aggregation *ex vivo* was observed at 1, 3 and 6, but at 9 hours after dosing in the young (Fig. 5). Duration of the effect of the drug was pro-

Fig. 3 Cumulative amount of urinary S-1452 and its metabolites after administration of 10 mg in the young ($n=8$, $\bigcirc$ — $\bigcirc$) and elderly ($n=8$, $\bullet$ — $\bullet$) Mean $\pm$ SD

Fig. 4 Ratio of metabolites [bisnor-(+)-S-145+tetranor-(+)-S-145] to S-1452 (total) in the values of $C_{\max}$ and AUC_{0-24} , and the 24-hour urinary excretion of these compounds after administration of 10 mg in the young ($n=8$) and elderly ($n=8$) Mean $\pm$ SD

Fig. 5 Platelet aggregation stimulated by U-46619 ($1\mu\text{M}$) after administration of 10mg S-1452 (●—●) and placebo (○—○) in the young ($n=8$) and elderly ($n=8$)
Mean $\pm$ SD
** : $p < 0.01$ compared with placebo

longed in the elderly, and the inhibition of platelet aggregation was detected at 9 hours after dosing.

There were no drug-related abnormalities in the physical examination, blood pressure, ECG or laboratory screening examinations. No adverse symptoms were recorded during this study.

Discussion

Plasma concentrations of S-1452 rapidly increased after oral dosing and the mean t_{max} was

0.7 hour in the young and 0.5 hour in the elderly. The AUC_{0-24} of S-1452 (total) + the metabolites, and the 24-hour urinary excretion of these compounds did not differ significantly between the young and elderly. These results suggest that the absorption of S-1452 is fast following oral dosing, and the amount of the drug absorbed from the gastrointestinal tract does not decrease with age.

It is generally accepted that the activity of the phase I hepatic drug metabolism is reduced or unchanged in the elderly, but the phase II pathway is unaffected¹⁾. S-1452 is extensively metabolized through the phase I (β -oxidation and hydroxylation) pathway⁶⁾. In the present study, the ratio of the metabolites (β -oxidation and hydroxylation) to unchanged compound in the C_{max} and AUC_{0-24} was lower in the elderly than in the young. In addition, there was no significant difference in the 24-hour urinary excretion of S-1452 and the metabolites between the young and elderly. These findings are compatible with the hypothesis that the metabolism of S-1452 through the phase I pathway decreases with age, which, in turn, might lead to the higher plasma concentration of S-1452 in the elderly. The decreased hepatic blood flow estimated by ICG-Kel might also be involved in the altered pharmacokinetic profiles of S-1452.

Previous study has shown that about 60% of the dose is excreted in the urine within the first 4 hours after administration of S-1452⁶⁾ which is consistent with the present data of the young ($62 \pm 12\%$) and elderly ($57 \pm 15\%$). Therefore, although the renal function estimated by the Cl_{cr} decreases with age, the urinary amount of S-1452 and the metabolites might not be diminished.

According to the results obtained from *in vitro* studies using human platelets, the antiag-

gregatory activity of S-1452 to collagen-or arachidonic acid-induced aggregation is qualitatively and quantitatively the same as that to U-46619-induced aggregation^{9,10}. ADP-induced aggregation consists of two steps. The first step which is induced through a specific receptor is not inhibited by S-1452, but the second step which is induced by TXA₂ produced during the first step process is completely inhibited by S-1452 with a concentration range similar to that required for other agents^{9,10}. Thus, U-46619 was chosen as a representative of other aggregating agents in this study.

The plasma unchanged S-1452 (active) rapidly decreased and could not be detected at 6 hours after oral dosing in the young and elderly. However, the inhibitory effect of S-1452 on platelet aggregation persisted more than 6 hours in this study. Such a long-lasting action could be, at least in part, explained by the extremely slow dissociation of S-1452 from platelet receptors¹¹. The pharmacodynamic effect of S-1452 was prolonged in the elderly. As shown in Figure 1, the plasma concentrations of unchanged S-1452 in the elderly tended to be higher than those in the young during the absorption phase. The higher S-1452 during the absorption phase might block more platelet TXA₂ receptors in the elderly. As the dissociation of S-1452 from platelet receptors is extremely slow¹¹, we think that the degree of TXA₂ receptor-blockade is sufficient to inhibit platelet aggregation at 9 hours after dosing in the elderly while the blockade of the receptor is negligible at the corresponding point in the young. Serum albumin in the elderly was significantly lower than that in the young in this study. As S-1452 is highly bound to human albumin (>97.2%, unpublished data), an increase in the unbound form of S-1452 might contribute to its prolonged effect in the elderly.

In summary, the present study showed that the plasma concentrations of S-1452 are higher and consequently, the inhibition of platelet aggregation is prolonged in the elderly. A previous study has shown that the bleeding time is slightly prolonged after 50 mg of S-1452 in young subjects¹². Although bleeding time was not determined in this study, it remains possible that bleeding time is further prolonged in the elderly. As S-1452 is generally prescribed on the basis of chronic use, repeated dosing studies are needed to assess the clinical relevance of these data.

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