Comparative Pharmacokinetic Study of Acetaminophen in Japanese and Han Chinese Individuals

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Abstract
Acetaminophen is widely used as a common analgesic, but toxic liver injury is a well known side effect of acetaminophen when used long-term. Compared to the clinical doses of acetaminophen used in Western countries and in China, the clinical doses used in Japan have been lower, and the drug potency is not high. The clinical doses of acetaminophen in Japan were increased in 2011, but the incidence of toxic liver injury remains to be investigated. In this study, the pharmacokinetics of acetaminophen were compared between Chinese and Japanese subjects, given that both are Asians and acetaminophen has been prescribed at high doses with an average of approximately 1,000 mg for many years in both countries. A single oral dose of acetaminophen 1,000 mg was administered to healthy Japanese and Chinese volunteers (8 participants each), and the pharmacokinetic parameters and urinary excretion rates were measured. No differences in pharmacokinetics of acetaminophen were observed between Japanese and Chinese subjects, and no differences in 24-h urinary excretion rates of unchanged acetaminophen and acetaminophen glucuronide as well as 4-acetaminophen sulfate and 3-cysteinyl acetaminophen were found. The pharmacokinetics of acetaminophen were similar in Chinese and Japanese subjects, and the risk of developing drug-induced toxic liver injury associated with an increase in acetaminophen dose is predicted to be comparable in the two ethnic groups.

Key words: acetaminophen, drug-induced liver injury, CYP2E1

Introduction
Acetaminophen is a widely used common analgesic, although the development of toxic liver injury is a well known side effect when used long-term1, Ingested acetaminophen follows a metabolic pathway in which approximately 85% of the drug is subjected to in vivo gluturonidation by uridine diphosphate-glucuronyltransferase (UGT), especially by UGT1A6, and sulfonation by sulfotransferase (SULT) 1A1 and SULT1A3. In addition, approximately 8–10% undergoes oxidation by cytochrome P-450 (CYP) 2E1. Particularly, N-acetyl-p-benzoquinone imine (NAPQI), which is an intermediate metabolite produced by metabolism of CYP2E1, has been reported to be a major cause of drug-induced liver injury (DILI)3. Excessive production of NAPQI because of a high level of chemical reactivity causes a depletion in glutathione (GSH) and strong binding with proteins, leading to cell death, thereby resulting in the development of DILI. Development of toxic DILI due to acetaminophen depends on the NAPQI generated through this series of metabolic activities, and may also depend on exposure. These could also be explained by data showing that toxic DILI often develops in heavy drinkers with induced CYP2E17.

The clinical doses of acetaminophen used in Japan have been lower than those used in Western countries and in China, and are considered to be effective. In 2011, the clinical doses in Japan were increased from a maximum of 300 mg per dose and a maximum dosage of 1,500 mg per day to a maximum of 1,000 mg per dose and a maximum dosage of 4,000 mg per day. However, the possibility of increase in risk of toxic DILI requires investigation. Ethnic differences in pharmacokinetics have been reported9, but no previous studies have compared the pharmacokinetics of acetaminophen and the above-mentioned series of metabolism between Japanese subjects and other ethnic groups. The Rumack-Matthew nomogram could be used to prognosticate the severity of possible liver toxicity after ingestion of acetaminophen10,11, but the development of DILI due to acetaminophen is dose-dependent and understanding the pharmacokinetics of acetaminophen is necessary. If the pharmacokinetics of acetaminophen in Japanese subjects show higher Cmax and AUC compared to other ethnic groups, the risk of DILI may also be higher. A previous study reported that the safety of high doses of acetaminophen does not differ between non-Japanese Asian and Western subjects12. In the present study, we compared the pharmacokinetics of acetaminophen between...
Chinese and Japanese subjects. We compared these two ethnic groups because both are Asians and acetaminophen has been prescribed at high dosages to Chinese subjects with an average of approximately 1,000 mg for many years. We also estimated the predicted frequency of DILI due to administration of high doses of acetaminophen.

**Methods**

**Participants**

Sixteen healthy male volunteers aged 20 years but <40 years (8 Japanese and 8 Chinese subjects) participated in this study. All subjects signed a written informed consent for participating in the study. In all participants, eligibility to participate in the study was determined by a screening test conducted by the physician in charge of the study. Subjects were excluded if they had a past history of anaphylaxis due to acetaminophen, peptic ulcer, blood abnormality or heavy alcohol consumption.

**Study design**

This study was not blinded and examined a single oral dose of acetaminophen.

**Drug administration**

For one week before the screening and one week before the study, as well as during the testing period, excessive physical exercise was forbidden in order to avoid possible impact on blood test results. On the day of study, the participants were instructed to refrain from reclining in a supine position for 4h after drug administration. The dose was determined taking into account the fact that the maximum dosage of acetaminophen in Japan was 500-700 mg per dose and 1,500 mg per day, while the maximum dosage in China was 600 mg per dose and 2,000 mg per day. From two weeks before oral administration of the test drug until discharge from the hospital, the participants were discouraged from using medications including nonprescription drugs, prescription drugs, Kampo drugs, and transdermal patches with systemic effects. They were also discouraged from taking health foods containing St. John’s Wort, unless the physician in charge of the study considered it necessary to use the foods, and unless the foods had no effect on pharmacokinetics. Ingestion of food and drinks was prohibited starting from 0 hour on the day of oral intake of the study drug, and drinking water was forbidden for a 4-h period starting from the time of administration of the drug, except during oral intake of the medication. In addition, food and drinks containing grapefruit, orange, and apple; alcohol-containing beverages; caffeine-containing beverages; and tobacco were also prohibited.

**Sample collection and processing, and analysis of the evaluation items**

Table 1 shows the time schedule used in this study. Approximately 7 mL of blood was collected immediately after drug administration, as well as at 15 min, 30 min, 1h, 1.5h, 2h, 4h, 6h, 8h, 10h, and 12h after drug administration. The samples were immediately cooled with ice and centrifuged (4℃, 3,000 rpm, 10 min), and plasma was then separated. The plasma samples were stored frozen at −70℃ or below until measurement of plasma drug levels. Urine was collected before administration of acetaminophen, as well as 0-4, 4-12, and 12-24h after administration of acetaminophen. Urine was harvested in a urine collection container, which was treated with ascorbic acid. The urine samples were stored in a cool and dark place and were sufficiently mixed. The total volume was then measured. Some of the collected urine was stored frozen at −70℃ or below until measurement of drug concentrations.

**Measurement of drug concentrations in plasma and urine and pharmacokinetic analysis**

The plasma and urinary levels of acetaminophen and acetaminophen glucuronide, 4-acetaminophen sulfate, 3-cysteinyl acetaminophen (a metabolite of NAPQI), and acetaminophen mercapturate were measured using LC/MC/MS methods. All specimens were measured at Shin Nippon Biomedical Laboratories, Ltd., Japan. All participants who were administered acetaminophen were analyzed. The pharmacokinetic parameters...
Table 2  Summary of pharmacokinetic parameters of acetaminophen (1,000 mg) in Japanese and Chinese subjects (n = 8 each)

<table>
<thead>
<tr>
<th>Pharmacokinetic parameter</th>
<th>Japanese subjects vs Chinese (Han-people) subjects</th>
<th>Acetaminophen</th>
<th>Acetaminophen glucuronide</th>
</tr>
</thead>
<tbody>
<tr>
<td>T_max, hr</td>
<td>0.38±0.13</td>
<td>0.41±0.13</td>
<td>0.41±0.13</td>
</tr>
<tr>
<td>C_max, µg/mL</td>
<td>18.8±5.88</td>
<td>16.3±4.1</td>
<td>21.56±7.34</td>
</tr>
<tr>
<td>AUC_{0-12}, µg·h/mL</td>
<td>55.8±11.94</td>
<td>56.2±10.7</td>
<td>136.5±32.1</td>
</tr>
<tr>
<td>T_{1/2}, hr</td>
<td>2.66±0.38</td>
<td>2.9±0.37</td>
<td>3.04±0.26</td>
</tr>
</tbody>
</table>

Values are means ± standard deviation.
The data were analyzed by t-test.

Statistics
All results were analyzed using JMP version 11.0 (SAS Institute Inc). Characteristic data and all pharmacokinetic parameters data were expressed as mean with standard deviation (SD). These data were analyzed using Student’s t-test.

Safety assessment
To evaluate participants’ safety, subjective and objective symptoms were observed, physical examinations were performed, and electrocardiography and clinical laboratory tests were also performed after administration of acetaminophen.

Ethical considerations
The study protocol was approved by Kitasato University Medical Ethics Organization (KEMEO) B10-30, G10-15. This study has been registered at the University Hospital Medical Information Network Clinical Trials Registry (UMIN-CTR; No. UMIN000003637). During preparation of case reports and handling of participants’ data, the privacy of the participants was taken into consideration and confidentiality was maintained using participant identification codes instead of the initials of the participants’ names. This also applied to participants who only underwent screening.

Results

Participant information
Of the 16 study participants, 8 were Japanese and 8 were Chinese subjects. The participants from each country were registered at the Kitasato University East Hospital Clinical Trial Management Center, and none of the participants dropped out until the end of the study.

No significant differences were found between the Japanese and Chinese participants with respect to age (30.3±6.3 vs. 27.1±3.3 years, p=0.236), height (172.1±7.2 vs. 173.9±7.8 cm, p=0.635), and body weight (69.1±11.9 vs. 67.2±9.3 kg, p=0.739).

Pharmacokinetic parameters
The plasma levels of acetaminophen increased rapidly after administration of the dose. Since almost no acetaminophen mercapturate was detected in plasma, analysis of this compound was not possible. There were no significant differences between Japanese and Chinese subjects in all the pharmacokinetic parameters including AUC_{0-12}, C_max, T_max and T_{1/2} (Table 2, Figures 1~4). There were no significant differences in urinary excretion rates of acetaminophen and all metabolites between Japanese and Chinese subjects (Table 3).

Safety evaluation
In terms of adverse events, among the 8 Chinese subjects who were administered acetaminophen, two participants developed internal bleeding in the left cubital fossa, and two other participants developed a nasal discharge. However, the events were mild and appeared to be unrelated to the test drug. No adverse events were observed in the 8 Japanese subjects.

Discussion
In this study, comparisons were carried out to determine whether the pharmacokinetics of acetaminophen differed between Japanese and Chinese subjects. For this purpose, Japanese and Chinese healthy volunteers were administered a single oral dose of acetaminophen, and the pharmacokinetics in each ethnic group were evaluated. No differences in age, body weight, and stature were found between the Japanese and Chinese subjects who participated in this study. According to a report published by the Ministry of Health, Labor, and Welfare in 2011, Japanese subjects aged between 18 and 29 years had a mean height of 170.8 cm and mean body weight of 65.4 kg, and according to
Figure 1  Time course of plasma acetaminophen concentration following a single dose of acetaminophen 1,000 mg in Chinese and Japanese healthy male subjects

The concentrations of metabolites were calculated as APAP equivalents.

Table 3  Cumulative amounts of drug excreted in urine (％ of dose)

<table>
<thead>
<tr>
<th>Drug</th>
<th>Japanese subjects (n=8)</th>
<th>Chinese (Han-people) subjects (n=8)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acetaminophen</td>
<td>2.1 ± 0.8</td>
<td>2.3 ± 0.8</td>
<td>p = 0.519</td>
</tr>
<tr>
<td>Acetaminophen glucuronide</td>
<td>49.8 ± 9.9</td>
<td>51.9 ± 12.5</td>
<td>p = 0.722</td>
</tr>
<tr>
<td>4-Acetaminophen sulfate</td>
<td>28.1 ± 8.5</td>
<td>25.7 ± 3.40</td>
<td>p = 0.498</td>
</tr>
<tr>
<td>3-Cysteinyl acetaminophen</td>
<td>3.77 ± 1.72</td>
<td>3.92 ± 3.01</td>
<td>p = 0.904</td>
</tr>
<tr>
<td>Acetaminophen mercapturate</td>
<td>1.01 ± 0.52</td>
<td>1.31 ± 1.23</td>
<td>p = 0.536</td>
</tr>
<tr>
<td>Total</td>
<td>84.6 ± 7.62</td>
<td>85.1 ± 9.13</td>
<td>p = 0.893</td>
</tr>
</tbody>
</table>

Values are means ± SD.
The data were analyzed by t test.
a report published by the General Administration of Sport of China in 2011, Chinese subjects aged between 25 and 29 years had a mean height of 170.7 cm and mean body weight of 68.7 kg. The Japanese participants in our study had a mean height of 171.9 ± 7.2 cm and mean body weight of 68.2 ± 11.9 kg, while the Chinese participants had a mean height of 173.7 ± 7.8 cm and mean body weight of 66.7 ± 9.3 kg. Therefore, the participants in our study were considered to have an average physique for Japanese and Chinese.

In terms of unchanged acetaminophen, acetaminophen glucuronide, 4-acetaminophen sulfate, and 3-cysteinyl acetaminophen levels, the pharmacokinetic parameters of acetaminophen were comparable in Japanese and Chinese subjects. Similar to the pharmacokinetic parameters, the urinary excretion rates of unchanged acetaminophen and its metabolites were also not different between Japanese and Chinese subjects.

The activity of UGT1A6 that catalyzes glucuronide conjugation of acetaminophen has been reported to be different between Westerners and Asians. However, Pei-Chieng et al. found no difference in UGT1A6 activity evaluated by expression of single-nucleotide polymorphisms (SNP) between Japanese and Chinese subjects, who are both East Asians. This is consistent with our result of no difference in metabolism of acetaminophen glucuronide between Japanese and Chinese subjects. Similar to the finding of UGT1A6, previous reports have also indicated no difference between Japanese and Chinese subjects with respect to SULT, which is an enzyme that catalyzes sulfate conjugation of acetaminophen. This finding is consistent with the result in the present study showing no difference in metabolism of acetaminophen sulfate between Japanese and Chinese subjects. As for CYP2E1, an enzyme that metabolizes acetaminophen to NAPQ and is considered to be the cause of DILI due to acetaminophen, slight differences in SNP have been reported between Japanese and Chinese subjects. However, in our study, the metabolism of 3-cysteinyl acetaminophen was not different between Japanese and Chinese subjects. Further, a lack of difference in physique, which is believed to have a significant impact on the blood levels of acetaminophen, may be one of the reasons for the absence of differences between Japanese and Chinese subjects in terms of the pharmacokinetic parameters of acetaminophen.

The results of previous clinical studies using intravenous formulation of acetaminophen in Japanese subjects and clinical studies using intravenous formulation of acetaminophen in Westerners have shown that Cmax and AUC tend to be slightly higher in Japanese than in Westerners. This result suggests that Japanese may be at higher risk of DILI than Westerners. High level of exposure is especially noted in Japanese subjects following increase of the approved dose. The risk of liver injury increases especially in cases where nonlinear pharmacokinetics is observed. However, the results of our previous study revealed linear pharmacokinetics of oral acetaminophen at doses of 300 mg, 600 mg, and 1,000 mg. Moreover, in a previous study conducted in Japan, patients with diseases or conditions such as osteoarthritis, cancer pain, and lower back pain were administered acetaminophen at doses of 2,400–4,000 mg/day. As a result, 7 of the 332 patients (approximately 2.1%) showed ALT levels three times higher than the reference values, and a causal relationship could not be ruled out in 3 of the patients (0.9%).

In a study reported by Kuffner et al., in which 1,039 osteoarthritis patients were administered 1,950–4,000 mg acetaminophen, ALT levels three times higher than the reference values were observed in 10 patients (approximately 1.0%), and causality could not be ruled out in 7 cases (approximately 0.7%). These results show similar tendencies to those found in previous studies conducted in Japan, and also suggest no significant difference in impact of large doses of acetaminophen on the occurrence of DILI between Westerners and Japanese, despite the physical differences between the two ethnic groups. To the best of our knowledge, no previous report had shown that severe DILI due to acetaminophen was more frequent in Chinese than in other ethnic groups. The absence of differences in pharmacokinetics for acetaminophen between Japanese and Chinese subjects, as shown in our study, also suggests that pharmacokinetically, administration of high doses of acetaminophen to Japanese subjects has no marked effect on the occurrence of DILI, compared to other ethnic groups.

Conclusions

The findings of this study indicate that the pharmacokinetics of acetaminophen are similar in Chinese and Japanese subjects, and that the risk of developing toxic DILI associated with an increase in dosage of acetaminophen may be predicted to be comparable in the two ethnic groups.

Conflict of Interest

All authors, except the corresponding author, have no conflict of interest to disclose. The corresponding author (Y. K.) received honoraria for medical specialist lectures from Showa Yakuhan Kako Co., Ltd.

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