Session 5B. Report 2.

THE POSSIBILITY OF PREDICTING TISSUE ACCUMULATION AFTER REPEATED DOSING USING A SINGLE-DOSE TISSUE DISTRIBUTION STUDY

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The guidance for repeated dose tissue distribution studies were established at the trilateral ICH meeting attended by the EU, US and Japan in 1994. Four circumstances under which repeated dose tissue distribution studies should be considered were described in the guidance. The first circumstance is described as 'when single dose tissue distribution studies suggest that the apparent half-life of the test compound (and/or metabolites) in organs or tissues significantly exceeds the apparent half life of the elimination phase in plasma and is also more than twice the dosing interval in the toxicity studies, repeated dose tissue distribution studies may be appropriate'.

This investigation surveyed how many of the tissue distribution studies conducted in Japan to date correspond to these conditions, and examined whether it is possible to predict tissue accumulation after repeated dosing using a single-dose tissue distribution study.

This paper was presented at the 5th Satellite Symposium of the Japanese Society of Toxicological Sciences on July 24, 1996 in Fukuoka.

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1. Repeated dose tissue distribution studies conducted in Japan

Repeated dose tissue distribution studies have been done for almost all drugs being developed using radioisotope labelled compounds (14C, 3H or 125I). The non-clinical safety evaluation subcommittee of JPMA investigated the number and contents of published reports involving repeated dose tissue distribution studies in Japan. There were 197 studies published in six related journals (Xenobio. Metabol. and Dispos. Dispos., Pharmacometrics, Clinical Report, Iyakuhin Kenkyu, Jpn. Pharmacol. Ther., Arzneim. Forsch.) between 1980 and 1993. Reports were collected from only a limited number of journals, thus findings might increase much further if studies reported in other journals or unpublished studies were added.

Then the studies reported in Xenobio. Metabol. and Dispos. from 1986 to 1993 were carefully reviewed. An index which indicates the accumulation tendency on repeated administration is represented as follows,

\[
\text{Accumulation ratio} = \frac{\text{tissue concn. at 24 hr after the final dose of reeated-dosing}}{\text{tissue concn. at 24 hr after single-dosing}}
\]
Currently, an accumulation ratio of 3 is considered the tentative critical value demonstrating an accumulation tendency. Among 51 compounds, 350 tissue specimens from 10 compounds showed an accumulation ratio above 3. Fig. 1 shows the incidence of an accumulation ratio above 3 in the 350 tissue specimens examined.

The order of probability for showing an accumulation ratio above 3 was: kidney, spleen, blood, liver, adrenal and skin.

2. Calculation of tissue concentration after repeated administration

The concentration in most tissues after a single administration could be fitted to a two-compartment equation,

\[ C_{\text{single}} = Ae^{-\alpha t} + Be^{-\beta t} - Ce^{-\gamma t} \]

Where \( C_{\text{single}} \) is the tissue concentration after a single administration, \( t \) is the time after administration and \( \alpha \), \( \beta \) and \( \gamma \) are the parameters for \( \alpha \) phase, \( \beta \) phase and absorption phase, respectively.

Then, the concentration after repeated administration can be predicted using the concentration parameters obtained as described above,

\[ C_{\text{repeated}} = A \left( \frac{1 - e^{-n\alpha \tau}}{1 - e^{-\alpha \tau}} \right) e^{-\alpha t} \]

\[ + B \left( \frac{1 - e^{-n\beta \tau}}{1 - e^{-\beta \tau}} \right) e^{-\beta t} - C \left( \frac{1 - e^{-n\gamma \tau}}{1 - e^{-\gamma \tau}} \right) e^{-\gamma t} \]

![Fig. 1](image-url) Occurrence frequency of specific accumulation in tissues by repeated administration of \(^3\)H- or \(^14\)C-labeled compounds. Tissues were surveyed from 51 \(^3\)H- or \(^14\)C-labelled compounds which were published in Xenobio. Metabol. and Dispos. from 1986 (Vol. 1, No. 1) to 1993 (Vol. 8, No. 2). [Reproduced from Mizojiri K. and Norikura R., Xenobio. Metabol. and Dispos. 11, 147 (1996)].
Where $C_{\text{repeated}}$ is the tissue concentration after repeated administration, $t$ is the dosing interval and $0 \leq t \leq \tau$.

Once steady state is attained the equation written above becomes as follows,

$$C_{\text{repeated}} = A \left( \frac{1}{1-e^{-\alpha \tau}} \right) e^{-\alpha t} + B \left( \frac{1}{1-e^{-\beta \tau}} \right) e^{-\beta t} - C \left( \frac{1}{1-e^{-\gamma \tau}} \right) e^{-\gamma t},$$

Then an accumulation index for the calculated concentration, accumulation factor, was defined as follows,

$$\text{Accumulation factor} = \frac{A \left( \frac{1}{1-e^{-\alpha \tau}} \right) e^{-\alpha t} + B \left( \frac{1}{1-e^{-\beta \tau}} \right) e^{-\beta t} - C \left( \frac{1}{1-e^{-\gamma \tau}} \right) e^{-\gamma t}}{A e^{-\alpha \tau} + B e^{-\beta \tau} - C e^{-\gamma \tau}}$$

If absorption and the $\alpha$ phase process finished before the next administration time, above equation is reduced to as follows,

$$\text{Accumulation factor} = \frac{1}{1-e^{-\beta \tau}}$$

Using this equation $t_{1/2\beta}$ of the tissue concentration at accumulation factor 3 is calculated as 41 hr.

3. **Comparison of accumulation factor (Calculated value) and accumulation ratio (observed value) of the tissue concentration after repeated dosing**

Tissue concentration after repeated dosing and accumulation factors could be calculated utilizing tissue concentration parameters of the single dose experiment according to the method previously described. Calculated accumulation factors were compared with accumulation ratios and were utilized to assess the predictability of accumulation.

Fig. 2 and Fig. 3 are the examples of calculations.

Fig. 2 shows the concentration of adipose tissue of compound A. This represents a case of comparably well agreement of the calculated and observed concentration after repeated dose. Fig. 3 indicates the concentration of submaxillary gland of compound B. This is a typical case of indicating a marked difference between observed and calculated concentration, and this suggests probable occurrence of unpredictable accumulation by repeated dose experiments.

The relationship between accumulation factor (calculated value) and accumulation ratio...
Fig. 3. Concentration of radioactivity in submaxillary gland after single or repeated oral administration of $^{14}$C-Compound B to rats (2 mg/kg/day, single and 21 days).
[Reproduced from Mizojiri K. and Norikura R., Xenbio. Metabol. and Dispos. 11, 147 (1996)].

Table 1. Indices for accumulation in tissue after administration of $^{14}$C-labeled compounds to rats.

<table>
<thead>
<tr>
<th>Range of accumulation ratio</th>
<th>1~3 (n=24)</th>
<th>3~5 (n=25)</th>
<th>5~7 (n=13)</th>
<th>7~ (n=11)</th>
<th>3~ (Total) (n=49)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tissue $t_{1/2} \beta$ at single dose (hr)</td>
<td>50.2±35.7</td>
<td>65.3±63.2</td>
<td>79.0±41.8</td>
<td>100.6±114.3</td>
<td>76.9±73.2</td>
</tr>
<tr>
<td>Accumulation factor (calculated value)</td>
<td>2.0±0.6</td>
<td>3.1±1.4</td>
<td>4.0±2.0</td>
<td>4.9±4.2</td>
<td>3.7±2.5</td>
</tr>
<tr>
<td>Accumulation ratio (observed value)</td>
<td>1.9±0.4</td>
<td>4.0±0.5</td>
<td>6.0±0.5</td>
<td>9.5±2.0</td>
<td>5.8±2.4</td>
</tr>
<tr>
<td>Ratio of tissue / plasma concn. ratio at 24hr (Repeade / Single)</td>
<td>1.3±0.4</td>
<td>2.2±0.8</td>
<td>3.0±1.4</td>
<td>4.2±1.8</td>
<td>2.9±1.8</td>
</tr>
</tbody>
</table>

[Reproduced from Mizojiri K. and Norikura R., Xenbio. Metabol. and Dispos. 11, 147 (1996)]

(observed value) and another index for accumulation are summarized in Table 1.

When the tissues from studies on 10 compounds were separated into four classes according to the accumulation ratio of 1-3, 3-5, 5-7 and more than 7, the $t_{1/2} \beta$ of these tissues after single dose showed means of 50.2, 65.3, 79.0 and 100.6 hr, respectively, indicating approximately proportional relationship between both values. Accumulation factors also increased with increase of accumulation ratios. Also, a tendency that large accumulation factors leading to great discrepancy between the accumulation factors and ratios was found. This great discrepancy suggests that unpredictable accumulation might happen with increase of the value of accumulation...
factor.

Tissue/plasma concentration ratio is also a useful indicator for assessment of accumulation tendency. In Table 1, repeated/single ratio of this ratio increased with increase of $t_{1/2} \beta$ of tissue concentration at single dose.

The possibility of prediction of tissue accumulation in repeated administration was investigated based on the published reports and it must be inevitably said that accumulation of the tissue concentration might become more remarkable and unpredictable when accumulation factor calculated from single dose tissue concentration becomes more than 3. Tissue distribution study in Japan have been mostly conducted using labelled compounds and this might be a serious problem because chemical form of the determined material was not clarified. However, this material could be evidently originated from dosing compound and this might accumulate unpredictably in tissues.

The tentative critical value of accumulation factor was proposed as 3 in this investigation. As described previously, $t_{1/2} \beta$ at accumulation factor 3 corresponds to 41 hr. This $t_{1/2} \beta$ value is almost similar to that of the critical value provided in the ICH guidance as should be considered a repeated dose tissue distribution study.