

Supplemental methods

Optimal timing analysis method

Assuming the one-compartment model for biological clearance, the percent injected dose per gram %ID/g of radiolabeled compounds A₁ and A₂ is expressed by the following respective equations:

$$(\%ID/g)_{A_1}(t) = \frac{A_{0_A_1} \exp(-\frac{\ln(2)}{T_{bA_1}}t)}{IAC_{0_A_1}}, \quad (s1)$$

$$(\%ID/g)_{A_2}(t) = \frac{A_{0_A_2} \exp(-\frac{\ln(2)}{T_{bA_2}}t)}{IAC_{0_A_2}}, \quad (s2)$$

where $A_{0_A_1}$ and $A_{0_A_2}$ are the initial activity concentrations, and T_{bA_1} and T_{bA_2} are HLs for the biological clearances of A₁ and A₂, respectively. Then, the activity concentration of A₁, $C_{A_1}(t)$, and that of A₂ with HL, $C_{A_2 \text{ with HL}}(t)$ are expressed by the following respective equations:

$$C_{A_1}(t) = A_{0_A_1} \exp(-\frac{\ln(2)}{T_{pA_1}}t) \exp(-\frac{\ln(2)}{T_{bA_1}}t), \quad (s3)$$

$$C_{A_2 \text{ with HL}}(t) = A_{0_A_2} \exp(-\frac{\ln(2)}{T_{pA_1}}t) \exp(-\frac{\ln(2)}{T_{bA_2}}t). \quad (s4)$$

The TIAC of A₁, $TIAC_{A_1}$, and that of A₂ with HL, $TIAC_{A_2 \text{ with HL}}$, were calculated by integrating equation (s3) and equation (s4), respectively:

$$TIAC_{A_1} = \int_{t_0}^{t_1} C_{A_1}(t) dt = \frac{A_{0_A_1}}{-\ln 2 \left(\frac{1}{T_{pA_1}} + \frac{1}{T_{bA_1}} \right)} \left[\exp \left(-\ln 2 \left(\frac{1}{T_{pA_1}} + \frac{1}{T_{bA_1}} \right) t \right) \right]_{t_0}^{t_1}, \quad (s5)$$

$$\begin{aligned}
TIAC_{A_2 \text{ with HL}} &= \int_{t_0}^{t_1} C_{A_2 \text{ with HL}}(t) dt \\
&= \frac{A_0 A_2}{-\ln 2 \left(\frac{1}{T_{pA_1}} + \frac{1}{T_{bA_2}} \right)} \left[\exp \left(-\ln 2 \left(\frac{1}{T_{pA_1}} + \frac{1}{T_{bA_2}} \right) t \right) \right]_{t_0}^{t_1}, \quad (s6)
\end{aligned}$$

where t_0 and t_1 are the start and end times of the evaluation period.

Next, the RAP coefficient at time t is expressed by the following equation, assuming

that $IAC_0_{A_1}$ is equal to $IAC_0_{A_2}$:

$$\text{RAP coefficient} = \frac{1}{\frac{(\%ID/g)_{A_2}(t)}{(\%ID/g)_{A_1}(t)}} = \frac{A_0 A_1 \exp\left(-\frac{\ln(2)}{T_{bA_1}} t\right)}{A_0 A_2 \exp\left(-\frac{\ln(2)}{T_{bA_2}} t\right)}. \quad (s7)$$

Finally, the optimal timing, Opt_t , was solved by inserting equations (s5) – (s7) into

equation (8) and taking the natural logarithms of both sides.

Infinite end time of the evaluation period

The evaluation period for optimal timing equation (8) could have an infinite end time. If

a one-compartment assumption for biological clearance holds for infinite time, the

optimal timing equation could be transformed as follows:

$$Opt_t = \frac{1}{-\ln 2 \left(\frac{1}{T_{bA_1}} - \frac{1}{T_{bA_2}} \right)} \ln \frac{\frac{1}{T_{pA_1}} + \frac{1}{T_{bA_2}}}{\frac{1}{T_{pA_1}} + \frac{1}{T_{bA_1}}}, \quad (s8)$$

where Opt_t is an optimal timing, T_{pA_1} is the physical HL of a target, T_{bA_1} is the biological

clearance HL of a target, and T_{bA_2} is the biological clearance HL of a reference. However,

verifiable data are rarely available for infinite time. In this paper, we proposed equation

(8) to calculate the optimal timing with a limited evaluation period, so that we could apply it even when the activity concentration data are available for only a short evaluation period. In fact, this was the case with our simulations of all datasets. In addition, an optimal timing formulated by equation (8) has limited application because of the assumption of the one-compartment model for biological clearance. This one-compartment assumption does not hold in the early stages of an evaluation period, when the uptake of radiolabeled compounds continues for a long time, as in equation (3). Therefore, in this case it is necessary to adjust the start time of the evaluation. This is an issue to be addressed in the future.