Assessment of the potency of a metabolite relative to the parent compound using a POP PK/PD model for the inhibition of a neurotransmitter re-uptake transporter in mice

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Background and Objectives

Drug X is a new CNS active compound which is currently under clinical development. X inhibits the re-uptake transporter of the neurotransmitter W and is mainly metabolised to Y, which is the only metabolite found in human plasma. Y shows significant *in vitro* activity, but has never been administered to any species, so its pharmacokinetic (PK) properties and also the *in vivo* activity are unknown. In order to investigate the *in vivo* contribution of the metabolite to the observed pharmacodynamic (PD) effect an animal study was performed to determine the potency and effect-time course of drug X and metabolite Y.

The objective of the data analysis was to develop a population PK/PD model in order to assess the potency of metabolite Y relative to parent drug X.

Methods

Study characteristics

Single doses of drug X or its metabolite Y were administered intravenously (0.3-10 mg/kg) or orally (1-20 mg/kg) to 132 female NMRI mice, 64 female NMRI mice were treated with placebo and served as control group. Samples for PK (X and Y) and PD were taken at 0.75, 1.5, 3, 16 and 17.5 h after administration. The PD effect was determined by the competitive inhibition of the re-uptake transporter of the neurotransmitter W by substance Z in mouse brain after administration of X or Y. Like X and Y, substance Z inhibits the re-uptake transporter W and was administered intravenously in a dose of 2.0 µCi as a tritium labelled molecule 45 minutes before the mice were decapitated. Selected areas of the mice brains were dissected and the amount of radioactivity per mg tissue was determined. Thus, the inhibition of the *in vivo* binding of drug X and metabolite Y by substance Z was determined.

Overall, 197 plasma concentrations (65 drug X, 132 metabolite Y) and 132 PD measurements, from 132 mice, were available for developing the PK/PD model.

Data analysis

The analyses were performed using the FO estimation method supplied by NONMEM version V, combined with graphical visualisation methods. Model development was performed in a sequential way, starting with a PK model for the metabolite after iv administration. Next, PK data from other administrations were added and the respective PK/PD models were built.

Results

Pharmacokinetics

Plasma concentration-time profiles of drug X and metabolite Y were best described by one compartment models with saturable Michaelis-Menten (MM) kinetics [Figure 2]. The schematic PK model is presented in Figure 1. Results of parameter estimates are listed in Table 1. Although nonlinear PK is present, PK parameters clearance and half-life could be derived within the linear PK range. Clearances were found to be high with 5.3 L/h/kg (X) and moderate with 1.9 L/h/kg (Y) which resulted in half-lives of 2.3 h (X) and 4.9 h (Y), respectively.

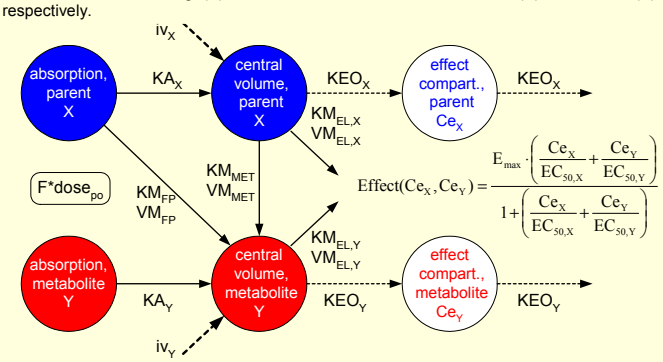

Figure 1. Schematic PK/PD model

Pharmacodynamics

The PD effect was best described by an extended Emax model which accounted for the competitive interaction of drug X and its metabolite Y [Figures 1, 2]. It was assumed that the efficacy (Emax) was equal for both compounds and that the maximum effect achievable by X and Y was a 100% inhibition of the neurotransmitter W re-uptake transporter. To account for the time delay in the PD, effect compartments were introduced in the model. Results of parameter estimates are listed in Table 1.

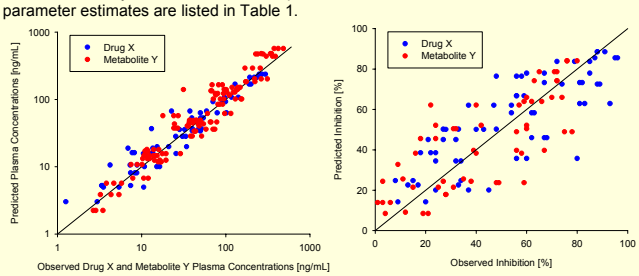

Figure 2. Observed versus predicted measurements. Split into PK (left panel) and PD (right panel) and separated into parent drug X (blue dots) and metabolite Y (red dots)

Table 1. Results of estimated parameters

Model Parameter		Estimate	Relative Standard Error %
Pharmacokinetic			
KA_X	[h^{-1}]	10 FIX	/
KA_Y	[h^{-1}]	5 FIX	/
$V_{central,\,X}$	[L/kg]	17.7	6.1
$V_{central,\,Y}$	[L/kg]	13.6	9.1
VM_{FP}	[mg/h/kg]	11.9	19.9
VM_{MET}	[mg/h/kg]	0.823	21.5
$VM_{EL,X}$	[mg/h/kg]	0.25	39.7
$VM_{EL,\,Y}$	[mg/h/kg]	0.506	14.4
$KM_{FP,\,MET,\,EL,X,\,EL,Y}$	[mg/kg]	3.59	21.0
F_X	[%]	87.2	5.1
F_Y	[%]	109	9.3
Pharmacodynamic			
E_{MAX}	[%]	100 FIX	/
$EC_{50,X}$	[ng/mL]	23.7	15.1
$EC_{50,Y}$	[ng/mL]	114.0	11.9
$KE0_X$	[h^{-1}]	0.555	29.0
$KE0_Y$	[h^{-1}]	0.878	24.3
Residual variability			
X, PK	[%]	35.7	17.1*
Y, PK	[%]	36.3	11.4*
X, PD	[%]	77.5	14.9*
Y, PD	[%]	89.3	15.3*

*given on the variance scale

Table 2. Estimated EC50 values of key models

Data	$EC_{50,X}$ [nM]	$EC_{50,Y}$ [nM]	$EC_{50,Y}$ / $EC_{50,X}$
Y iv*	n.a.	318.4	n.a.
Y iv + X iv*	76.2	309.2	4.1
Y iv + X iv and po	81.4	340.8	4.2
Y iv and po + Y iv and po	72.3	363.1	5.0

*no effect compartment necessary
n.a.: not applicable

Discussion

Pharmacodynamic investigations revealed a 4.1 to 5.0 fold higher *in vivo* potency of drug X in comparison to metabolite Y regarding the inhibition of the neurotransmitter W re-uptake transporter in mice [Table 2]. As metabolite Y shows in humans approximately 60% lower steady state concentrations compared to drug X suggest that the contribution of the active metabolite Y to the overall effect is probably low.

Comparison of EC50 values of iv data and iv plus po data indicated that no additional active metabolites were built during the first-pass metabolism of drug X [Table 2]. Time delay between PK and PD, described by effect compartments, might be caused by distribution processes of the molecules between plasma and the CNS. However, as drug X is intended for chronic treatment, distribution processes play a minor role.

Pharmacokinetics of drug X and its metabolite Y exhibited non-linearity which had never been noticed before in any species treated. Simulations performed with parameters obtained from this data analysis showed that MM kinetics have only an impact on high concentrations caused by high doses administered in this trial [Figure 3]. However, non-linearity in human PK is not expected to occur within the effective plasma concentration range.

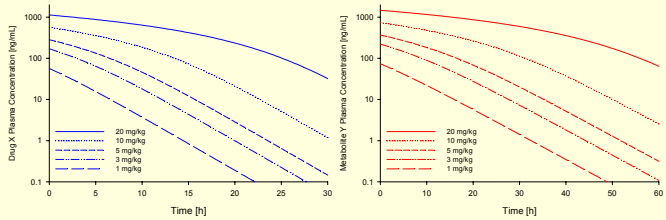

Figure 3. Simulated plasma concentration-time profiles after bolus infusion of different doses. Left panel (blue lines) shows the parent drug X, right panel (red lines) shows the metabolite Y.

Conclusion

- A model was successfully developed describing simultaneously the PK and PD data of drug X and its metabolite Y.
- Observed non-linearity in PK might be caused by high doses administered.
- EC50 values did not indicate generation of further active first-pass metabolites.
- Drug X showed a 4.1 to 5 fold higher *in vivo* potency in mice in comparison to its metabolite Y with respect to the inhibition of the neurotransmitter W transporter.
- A five fold lower potency of metabolite Y and approximately 60% lower steady-state plasma concentrations of Y in humans compared to drug X suggest that the contribution of the active metabolite Y to the overall efficacy is probably low.