PXB-mice with new donor hepatocytes: Reproducible animal model for prediction of human pharmacokinetic parameters

Teruki Hamada, Noriyasu Sano, Kazunobu Aoyama, Yasuyuki Debori, Yukiko Watanabe, Shuntarou Tsuchiya, Mikio Shirasaki and Nobuyuki Amano

Drug Disposition & Analysis, Research Division, Axcelead Drug Discovery Partners, Inc.

Purpose

We clarified whether pharmacokinetic (PK) data of test compounds in chimeric mice with humanized liver (PXB-mice) from new donor were comparable with those in published data using PXB-mice with previous donor hepatocytes.

Moreover, we investigated predictability of human PK parameters of the compounds, and evaluated a formation clearance of major human metabolite of midazolam in both PXB-mice and severe combined immuno-deficiency (SCID) mice.

Materials and Methods

Animals: **PXB-mice (PhoenixBio Co., Ltd.; with Lot A hepatocytes) and SCID mice (Charles River Laboratories Japan, Inc.)**

Dosing regimen: **Intravenous (i.v.) 0.1 or 0.3 mg/kg, oral (p.o.) 1 mg/kg**
All animal experiment protocols were approved by the Institutional Animal Care and Use Committee of Shonan Health Innovation Park.

Bioanalysis: **Concentrations in plasma and urine by LC/MS/MS**

PK analysis: **Non-compartmental analysis**
CLr = Ae,u / AUCp and CLh = CLtotal – CLr
where CLr: renal clearance, CLh: hepatic clearance, Ae,u: cumulative amount in urine

Prediction of human CLtotal and Vd(ss): **Single-species allometric scaling method (SSS) with PXB-mouse data**
Body weight: 60 kg (humans) and 0.02 kg (PXB-mice). Exponent for SSS: 0.75 (CLtotal) and 1 (Vd(ss)). Assumption of "fu,p (PXB-mice) = fu,p (humans)" was adopted.

Table I Characteristics of test compounds

Compound	Major biomolecule in humans involved in compound disposition
Antipyrine	Non-specific P450
Diclofenac	CYP2C9, UGT
Midazolam	CYP3A4, 3A5
BIBX1382	Aldehyde oxidase (AO)
Pitavastatin	OATP1B1, CYP2C9, UGT
Olmesartan	OATP1B1, 1B3, MRP2

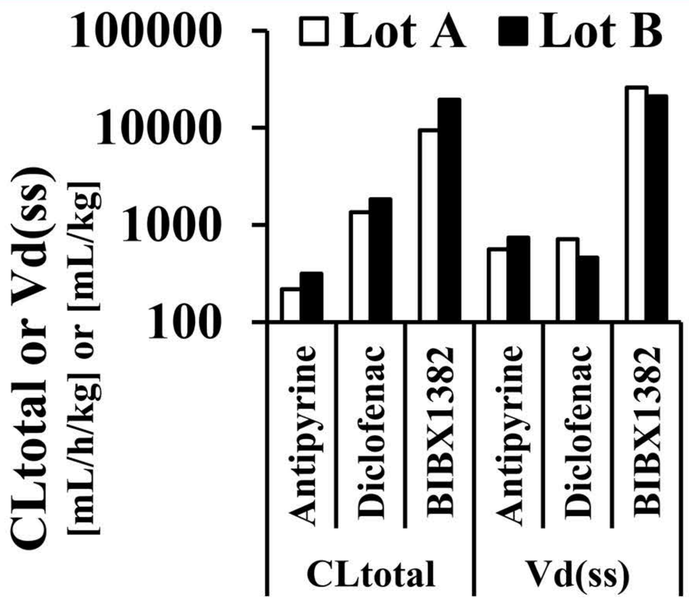
Table II Characteristics of donors for transplanted human hepatocytes to PXB-mice

Characteristics	Lot A	Lot B	Lot C
Hepatocyte lot number	IVT-JFC (new lot)	BD195 (previous lot)	BD85 (previous lot)
Ethnic group	Caucasian	Hispanic	African-American
Gender, Age	Male, 1y	Female, 2y	Male, 5y

Results

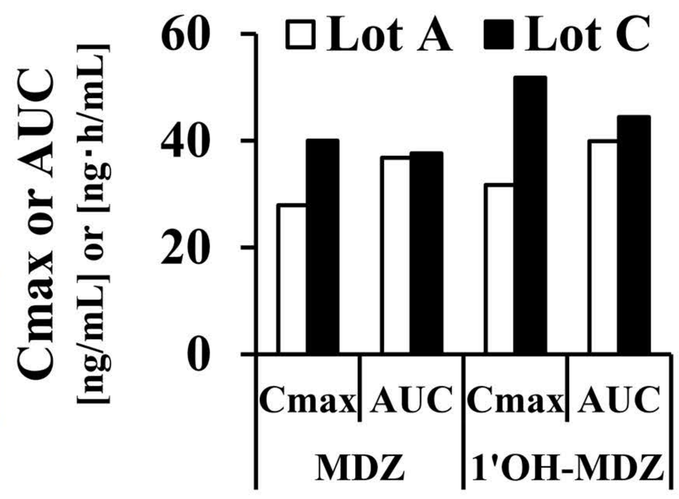
Reproducibility of PK parameters

Fig. 1. Reproducibility of PK parameters in PXB-mice with Lot A. (Comparison with PXB-mice with Lot B[1])



CLtotal and Vd(ss) in PXB-mice with Lot A: ≦ 2-fold of mice with Lot B

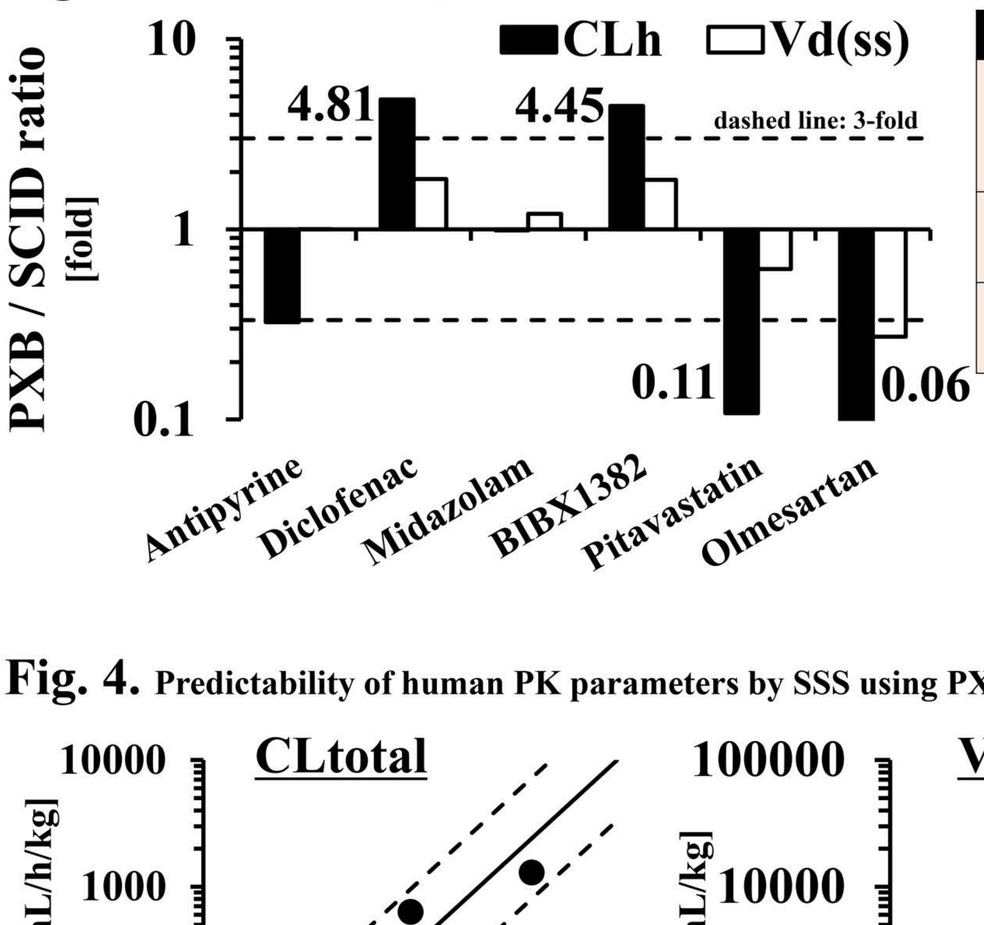
Fig. 2. Reproducibility of PK parameters of midazolam (MDZ) and one of its metabolites, 1'-hydroxymidazolam (1'OH-MDZ), in PXB-mice with Lot A after oral dosing of MDZ. (Comparison with PXB-mice with Lot C[2])



Cmax and AUC in PXB-mice with Lot A: ≦ 2-fold of mice with Lot C

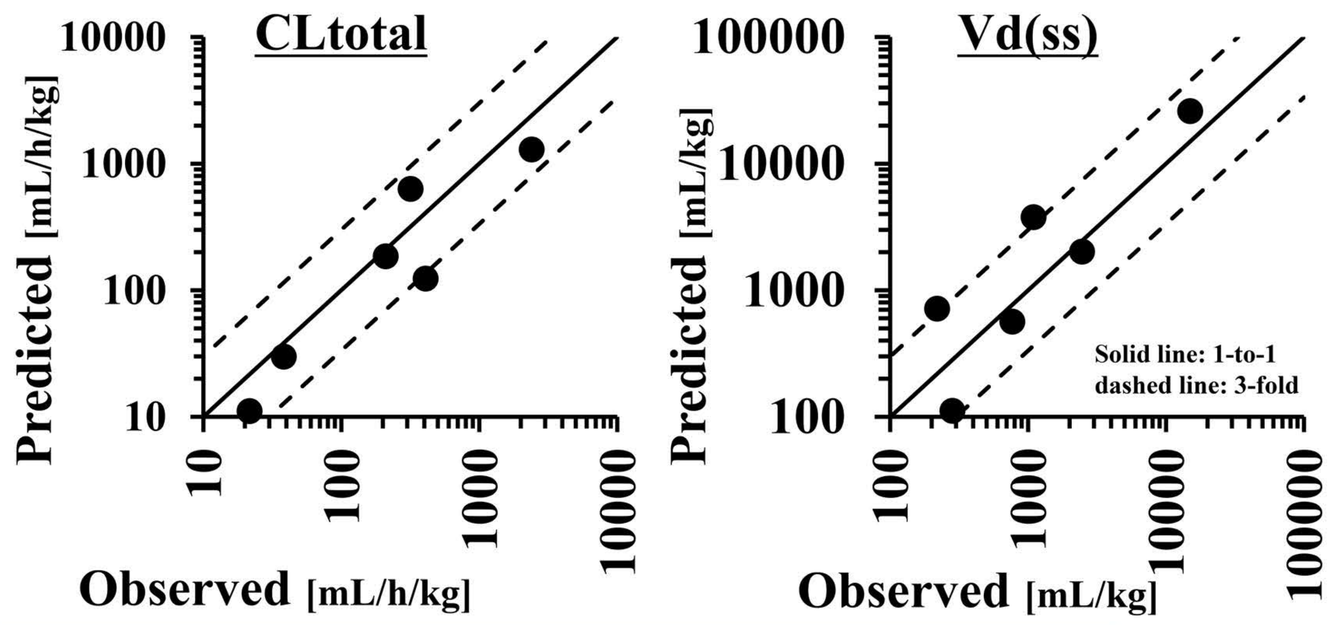
Predictability of human PK parameters and major metabolites

Fig. 3. Comparison of PK parameters between PXB-mice with Lot A and SCID mice.



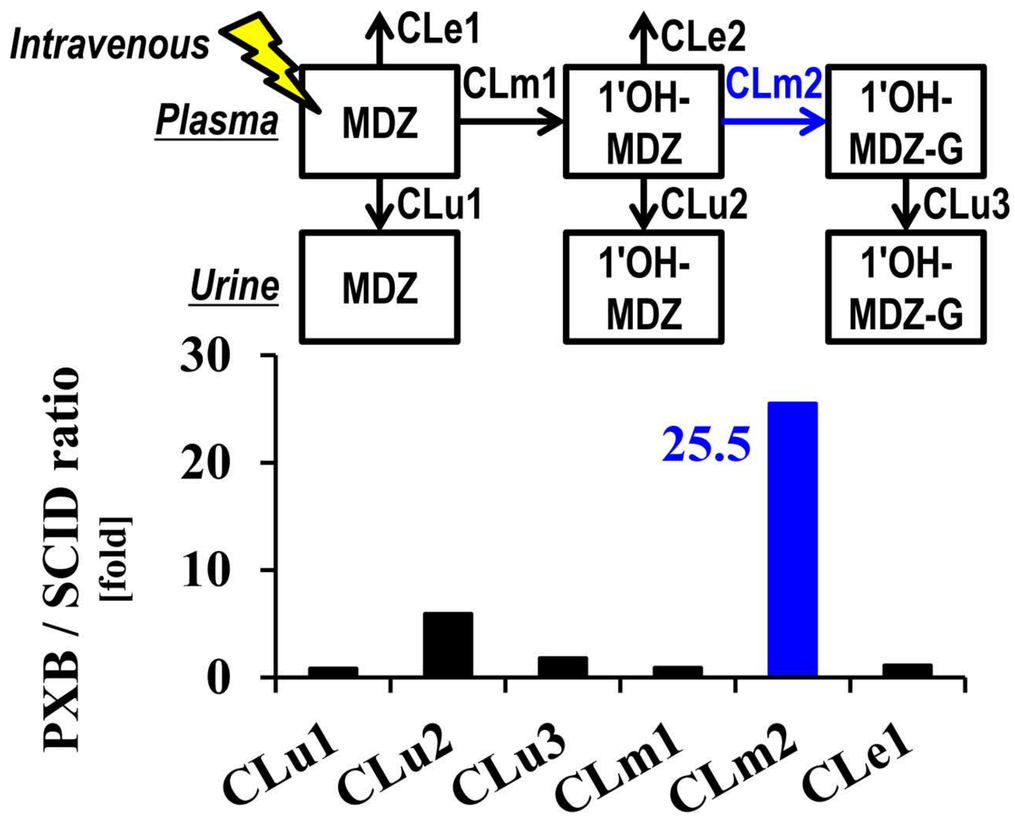
Compound	Results	Published data
Diclofenac	Metabolic clearance (P450, UGT): PXB > SCID	Oxidation & glucuronidation clearance (liver microsome): Human > Mouse[3,4]
BIBX1382	Metabolic clearance (AO): PXB > SCID	AO metabolism activity (liver S9): Human > Mouse[5]
Pitavastatin	Hepatic uptake clearance (OATP): PXB < SCID	OATP transport activity (in vivo clearance): OATP1B1, 1B3-humanized mice < Wild type[6]

Fig. 4. Predictability of human PK parameters by SSS using PXB-mice with Lot A.



Predicted human CLtotal and Vd(ss) : ≦ 3-fold of the clinical data
The results were consistent with those in PXB-mice with Lot B[1].

Fig. 5. Disposition model (upper) and clearance ratios between PXB-mice with Lot A and SCID mice (lower) from MDZ to 1'OH-MDZ glucuronide (1'OH-MDZ-G).



CLm2: PXB-mice >> SCID mice
The results were consistent with those in PXB-mice with Lot C[2].

Conclusion

Effect of inter-lot difference of human hepatocytes in PXB-mice on PK parameters was limited.

PXB-mice are useful to predict human PK parameters and to evaluate unique or major human metabolites.

COI disclosure information

PXB-mice were provided from PhoenixBio Co., Ltd. free of charge. We have no other financial relationship to disclose for our presentation contents.

References:
[1] Miyamoto M, et al. Xenobiotica. 2017;47(12):1052-1063.
[2] Samuelsson K, et al. Xenobiotica. 2012;42(11):1128-37.
[3] Bogaards JJ, et al. Xenobiotica. 2000;30(12):1131-52.
[4] Fujiwara R, et al. Drug Metab Pharmacokinet. 2018;33(1):9-16.
[5] Crouch RD, et al. Xenobiotica. 2018; 48(3):219-231.
[6] Salphati L, et al. Drug Metab Dispos. 2014;42(8):1301-13.