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A myth of the well-stirred model: Is the well-stirred model good for high clearance drugs?

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ABSTRACT

Understanding the rationale of the well-stirred model (WSM), borrowed from chemical engineering, has been ongoing through the history of pharmacokinetics (PK) as an independent discipline. Extensive arguments around the WSM and 1977′s lidocaine data re-emerged recently. It was proposed that Pang and Rowland's lidocaine data analysis was confounded by four intermingled confounding factors which may lead to contradictory conclusions or inconclusive dilemma. This re-visit of 1977′s lidocaine data analysis was challenged by Pang and coauthors. This commentary is our responses to their comments focusing on the lidocaine data analysis and the IVIVE by the WSM. In addition, the disadvantage of applying the well-stirred model in drug-drug interaction (DDI) prediction and a theoretical dilemma in the commonly used whole-body physiologically based pharmacokinetic (PBPK) models were discussed.

1. Introduction

It has been well recognized that the well-stirred model (WSM) gives poor *in vitro-in vivo extrapolation* (IVIVE) for high clearance drugs (Chiba et al., 2009; Watanabe et al., 2009) (note: drugs with hepatic extraction ratios ($E_{h,B}$) greater than 0.8 are considered as high clearance drugs while the ones with $E_{h,B}$ less than 0.8 are low-to-moderate clearance drugs in this paper). However, 1977′s lidocaine data suggested that the WSM is better than the parallel-tube model (PTM) when the lidocaine $E_{h,B}$ was higher than 0.99 (Pang and Rowland, 1977). A re-analysis was performed to clarify the discrepancy (Dong and Park, 2018). The results showed: (a) the WSM is very similar to the PTM and dispersion model (DM) when predicting $E_{h,B}$ or clearance for low-to-moderate and high clearance drugs, (b) the PTM is better than the WSM when estimating intrinsic clearance ($CL_{h,int}$) for high clearance drugs, (c) neither WSM nor PTM is recommended to predict hepatic availability ($F_{h,B}$) for high clearance drugs (Dong and Park, 2018). Recently, Pang and coauthors (Pang et al., 2019) challenged the re-analysis. Herein, we would like to respond to Pang and coauthor's arguments focusing on the lidocaine data analysis and the IVIVE by the WSM. In addition, the disadvantage of applying the well-stirred model in drug-drug interaction (DDI) prediction and a theoretical dilemma in the commonly used whole-body physiologically based pharmacokinetic (PBPK) models were discussed.

2. Four intermingled factors

The concerns on 1977′s lidocaine data analysis was not due to a single factor of sensitivity or relative error but four intermingled factors (Dong and Park, 2018). The four intermingled factors may cause unstable comparison and depending on the selection of the control condition, could lead to different conclusions and sometimes inconclusive result dilemma, such as for diazepam and diclofenac as shown in the previous paper (Dong and Park, 2018). In addition, regardless of the relationship between the WSM and PTM, the simulation by the DM is expected to be closer to the observed values than the WSM, given that the WSM is an approximation or extreme case of the DM. Yet, the conclusion drawn using 1977′s lidocaine data is the opposite, as shown in Fig. 1A. The four intermingled factors incorporating the comparison with the simulations by the DM were further discussed in the Appendix A in the Supplementary Materials following the same strategy we reported previously (Dong and Park, 2018). Fig. 1A is a result of: on the WSM side, it has high relative error for estimating $CL_{h,int}$ and predicting $F_{h,B}$; on the PTM and DM side, they have too high sensitivity for predicting $F_{h,B}$. And the instability issue was amplified by using different back-calculated $CL_{h,int}$ values for simulation. All the four factors intermingled together leading to an unstable and less reliable comparison of the WSM and the PTM/DM.

3. Comparison by IVIVE of lidocaine

Pang and coauthors challenged the comparison approach of using the same *in vitro* $CL_{h,\ int}$ to predict *in vivo* $E_{h,B}$ with the WSM and PTM. In fact, this is one of the most common practices of IVIVE in both academia and industry.

Today's IVIVE is not perfect yet but it does not mean it always causes misprediction, especially underprediction. The commonly observed underprediction occurs more often for drugs as substrates of transporters, non-cytochrome P450 (non-CYP) enzymes, or having high plasma protein binding (Bowman and Benet, 2016, 2019; Poulin et al., 2012). It is critical to apply the knowledge of elimination routes and the

Abbreviations: AUC, area under the curve; BDDCS, biopharmaceutical drug disposition classification system; CYP, cytochrome P450; DDI, drug-drug interaction; DM, dispersion model; ECCS, extended clearance classification system; IVIVE, *in vitro-in vivo* extrapolation; non-CYP, non-cytochrome P450; PBPK, physiologically based pharmacokinetics; PK, pharmacokinetics; PTM, parallel-tube model; WSM, well-stirred model.

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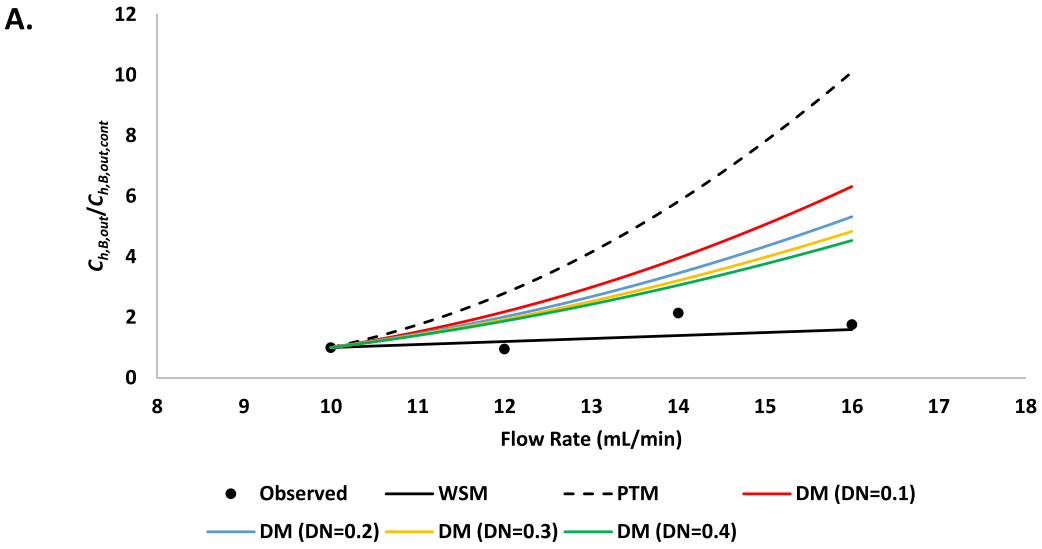

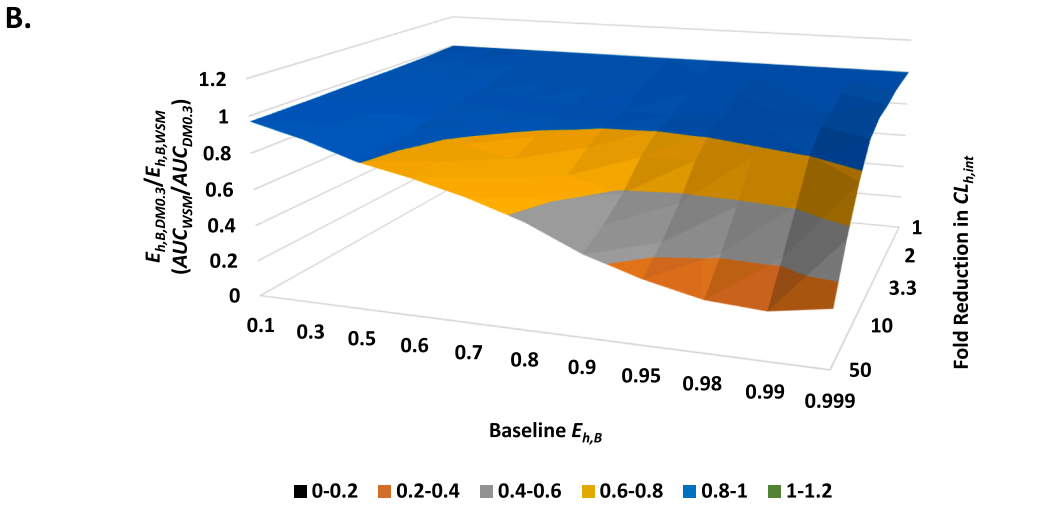

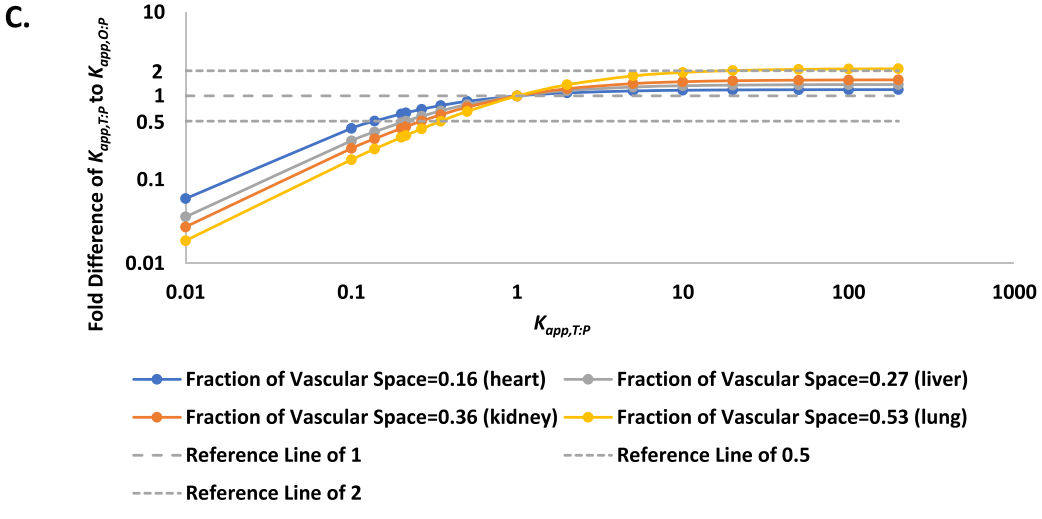

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Fig. 1. (A). The observed and simulated output lidocaine concentrations ($C_{h,B,out}$) at different flow rates divided by observed output lidocaine concentrations at the control flow rate of 10 mL/min ($C_{h,B,out,cont}$) (Pang and Rowland, 1977). (B). Drugs with baseline $E_{h,B}$ from 0.1 to 0.999 upon 1 to 50-fold reduction in their $CL_{h,int}$. It was assumed that: i) overall bioavailability is 1 (intravenous administration); ii) hepatic elimination is the only elimination route; iii) $f_{u,B}$ and $Q_{h,B}$ are not changed. $E_{h,B,WSM}$ is calculated by the WSM with the reduced $CL_{h,int}$ (baseline $CL_{h,int}$/fold reduction. Baseline $CL_{h,int}$ was estimated by the WSM from the baseline $E_{h,B}$). Similarly, $E_{h,B,DM0.3}$ is calculated by the DM ($D_N = 0.3$) with the reduced $CL_{h,int}$ (baseline $CL_{h,int}$/fold reduction. Baseline $CL_{h,int}$ was calculated by the DM from the same baseline $E_{h,B}$). (C) The fold difference of $K_{NE,app,T:P}$ to $K_{NE,app,O:P}$ (calculated by $\frac{K_{app,T:P}}{K_{app,O:P}}$) when $K_{NE,app,T:P}$ is in the range of 0.01∼200 and $R_{B:P}$ is assumed as 1. Heart, fraction of vascular space of 0.16, blue; liver, fraction of vascular space of 0.27, gray; kidney, fraction of vascular space of 0.36, orange; lung, fraction of vascular space of 0.53, yellow. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

involvement of transporters and non-CYP enzymes before the effort of evaluating the IVIVE using traditional metabolism-based *in vitro* assays such as microsome and suspended hepatocyte assays. In contrast, one may have higher confidence on establishing metabolism prediction for drugs metabolized by CYP enzymes with high passive permeability and low to moderate plasma protein binding. Otherwise, the evaluation of the goodness of IVIVE may be confounded. In the case of lidocaine, it is a biopharmaceutical drug disposition classification system (BDDCS) class I or extended clearance classification system (ECCS) class II drug with high passive permeability. It is primarily metabolized by CYP enzymes. Its unbound fraction in the perfusate ($f_{u,pf}$) in 1977′s lidocaine study was 0.95 (Pang and Rowland, 1977). And, its plasma protein binding in rat, monkey, and human are 0.43, 0.39, and 0.33, respectively (Lombardo et al., 2013). Thus, lidocaine is one of the drugs one may have higher confidence on its IVIVE.

In addition, if the *in vitro* and *in vivo* $CL_{h,int}$ showed good correlation by the PTM and DM model while there is significant underprediction by the WSM for a high clearance drug, such as observed in lidocaine, an IVIVE may be established and the mismatch by the WSM could be due to the overestimation issue of the WSM itself which has been well discussed (Chiba et al., 2009; Dong and Park, 2018).

4. The well-stirred model for DDI

The WSM has distinct mathematical features of the relative error and sensitivity when estimating *in vivo* $CL_{h,int}$ for low-to-moderate and high clearance drugs (Dong and Park, 2018), while it is common to use back-calculated *in vivo* $CL_{h,int}$ from *in vivo* clearance by the WSM. However, it was shown in Fig. 1B that the back-calculated *in vivo* $CL_{h,int}$ from *in vivo* clearance using the WSM may lead to underprediction (<0.8) of area under the curve (AUC) for drugs with baseline $E_{h,B}$ greater than 0.5 upon larger than 3.3-fold reduction in their $CL_{h,int}$ compared with using the DM ($D_N = 0.3$). Drugs with very high baseline $E_{h,B}$ may be immune from the < 0.8-fold of difference due to $CL_{h,int}$ reduction. For example, the fold difference was 0.91 for drugs with baseline $E_{h,B}$ of 0.999 upon 5-fold reduction in $CL_{h,int}$. This is because the reduced $CL_{h,int}$ is still very high making the drugs as high extraction ratio drugs whose clearance is dominated by the blood flow rate. In addition, it is not a problem when predicting the AUC reductions due to $CL_{h,int}$ induction using *in vivo* $CL_{h,int}$ back-calculated with the WSM, as shown in Fig. S1. Albeit the limitation of the above analysis performed under the assumption of steady state, the preliminary simulation results suggested that the use of WSM for back-calculating $CL_{h,int}$ in physiologically based pharmacokinetic (PBPK) modeling warrants further investigation on its potential impact on the simulation in the scenarios when the drug $E_{h,B}$ varies over a large range across the category of low-to-medium and high clearance, especially due to $CL_{h,int}$ reduction (e.g., drug-drug interaction (DDI), polymorphism, disease effect, pediatrics, etc.).

5. The theoretical dilemma in PBPK models relating to concentration calculation

Population based PBPK is critical to incorporate the variabilities in clearance prediction. However, a theoretical dilemma is found from the commonly used PBPK models in their mass balance when the vascular spaces of each organs are lumped to the arterial and venous blood reservoirs.

Herein we briefly discuss the well-stirred non-eliminating organ model only (the detailed theoretical derivations including the liver model are presented in Appendix C). The following equation is used in the traditional PBPK models (Jones and Rowland-Yeo, 2013; Peters, 2012).

$$V_{NE,T} \cdot \frac{dC_{NE,T}}{dt} = Q_{NE,B} \cdot \left(C_{NE,B,in} - C_{NE,B,out}\right) \quad (1)$$

where $C_{NE,B,in}$ and $C_{NE,B,out}$ are the blood concentration of drug entering and leaving the non-eliminating organ, respectively; $C_{NE,T}$ is the tissue concentration of the non-eliminating organ; $V_{NE,T}$ is the tissue volume (which excludes the vascular space) of the non-eliminating organ; $Q_{NE,B}$ is the blood flow rate of the non-eliminating organ.

To relate $C_{NE,B,out}$ and $C_{NE,T}$ and reduce the number of unknown variables, Eq. (2) was proposed under the assumption of perfusion limited transport (Jones and Rowland-Yeo, 2013; Jones et al., 2006; Peters, 2012)

$$C_{NE,B,out} = \frac{C_{NE,T}}{K_{NE,app,T:P}/R_{B:P}} = \frac{C_{NE,T}}{K_{NE,app,T:B}} \quad (2)$$

where $K_{NE,app,T:P}$ and $K_{NE,app,T:B}$ are the apparent partition coefficient of drug between the tissue and the emergent venous plasma and blood of the non-eliminating organ, respectively; $R_{B:P}$ is the blood to plasma ratio. Here, the tissue concentration in the definition of $K_{NE,app,T:P}$ is "a concentration of drug in a tissue outside of the blood perfusing it" (Rodgers et al., 2005). In other words, the blood concentration is excluded in the calculation of the tissue concentration.

However, the total change rate in the amount of drug in the non-eliminating organ should include both vascular and tissue spaces as

$$\frac{dA_{NE}}{dt} = \frac{dA_{NE,B}}{dt} + \frac{dA_{NE,T}}{dt} = V_{NE} \cdot \frac{dC_{NE}}{dt} = Q_{NE,B} \cdot \left(C_{NE,B,in} - C_{NE,B,out}\right) \quad (3)$$

where A_{NE}, $A_{NE,B}$, and $A_{NE,T}$ are the total drug amount (includes both vascular and tissue spaces), the drug amount in the vascular space and the drug amount in the tissue space in the whole non-eliminating organ, respectively; C_{NE} and V_{NE} are the average organ concentration and total volume of the non-eliminating organ.

Comparing Eq. (1) with Eq. (3), the right-hand side of these two equations are exactly the same while the left-hand sides are different. In the traditional non-eliminating organ model, $V_{NE,T} \cdot \frac{dC_{NE,T}}{dt}$ is not the total change rate in the amount of drug in the whole non-eliminating organ, which includes both vascular and tissue spaces. Eq. (1) does not hold the principle of mass conservation except when all the change rates are zero under steady state. If the traditional definition of $C_{NE,T}$ is a misphrasing but in practice the average concentration of the whole organ in the traditional PBPK modeling is used, then the use of Eq. (2) is inappropriate, especially when $K_{NE,app,T:B}$ is not 1 (the average drug concentration of the whole non-eliminating organ C_{NE} is not equal to $C_{NE,T}$). This is because the blood concentration is not being taken into account for calculating the average concentration of the whole organ. $K_{NE,app,O:P}$, the

ratio between C_{NE} and $C_{NE,B,out}$, rather than $K_{NE,app,T:P}$, is needed for solving Eq. (3).

$$K_{NE,app,O:P} = K_{NE,app,T:P} \cdot \frac{V_{NE,T}}{V_{NE,B} + V_{NE,T}} + R_{B:P} \cdot \frac{V_{NE,B}}{V_{NE,B} + V_{NE,T}} \quad (4)$$

where $V_{NE,B}$ is the blood volume of the non-eliminating organ.

Eq. (4) and Fig. S4 suggested that the difference between $K_{NE,app,O:P}$ and $K_{NE,app,T:P}$ is larger in richly perfused organs such as heart, liver, kidney and lung with fraction of vascular space of 0.16, 0.23~0.36, 0.14~0.27, 0.53, respectively (Table S1). The simulation results showed larger than two-fold difference in drugs with heart, kidney, liver, and lung $K_{app,T:P}$ less than 0.138, 0.265, 0.213, and 0.346 assuming $R_{B:P}$ as 1 (Fig. 1C). For example, ceftazidime showed $K_{NE,app,T:P}/K_{NE,app,O:P}$ of 0.09 for heart, 0.12 for liver, and 0.13 for lung in the rat (Table S2; $K_{NE,app,T:P}/K_{NE,app,O:P}$ of 0.88 for kidney due to the relatively high observed $K_{NE,app,T:P}$ of 0.54 greater than 0.265; $K_{NE,app,T:P}/K_{NE,app,O:P}$ of 0.15 for heart, 0.20 for liver, and 0.21 for lung assuming $R_{B:P}$ as 0.55).

As the vascular space should be taken into account in each organ model to maintain its mass balance, the blood reservoir compartments are removed to avoid doubling the vascular space in the whole-body PBPK model. The lung compartment becomes

$$V_{lung} \cdot \frac{dC_{lung}}{dt} = \sum Q_{i,B} \cdot C_{i,B,out} - Q_{lung,B} \cdot C_{lung,B,out} \quad (5)$$

where C_{lung}, V_{lung}, and $Q_{lung,B}$ are the average organ concentration, the total volume, and the blood flow rate of the lung (which is equal to the cardiac output), respectively; $C_{i,B,out}$ and $Q_{i,B}$ are the blood concentration of drug leaving the *i*th organ and the blood flow rate of the *i*th organ, respectively. Therefore, it is critical to incorporate the "peripheral sampling site" model (Musther et al., 2015) to simulate the sampled blood/plasma concentration.

6. Conclusions

Regardless of the relationship between the WSM and PTM, the WSM is an extreme case of the DM according to Roberts and Rowland's theory. However, why does lidocaine data show the WSM to be better than the DM? How to explain the contradictory or inconclusive comparison of the WSM and PTM using diazepam and diclofenac data by applying 1977′s lidocaine data analysis method? These were not addressed in the commentary (Pang et al., 2019).

In contrast, the IVIVE approach showed the PTM and DM were better than the WSM for lidocaine, diazepam, and diclofenac by avoiding the relative error issues of the WSM and stability issues of the PTM and DM when predicting $F_{h,B}$ (Dong and Park, 2018). Caution may be needed when using the WSM for building IVIVE for high clearance drugs when predicting $F_{h,B}$ and estimating $CL_{h,int}$ and when using WSM to back-calculate $CL_{h,int}$ for drugs with $E_{h,B}$ higher than 0.5 for scenarios such as simulating inhibition-mediated DDI. In addition, the theoretical dilemma in mass balance of the PBPK models warrants further investigation.

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Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.ejps.2022.106134.

References

Bowman, C.M., Benet, L.Z., 2016. Hepatic clearance predictions from in vitro–in vivo extrapolation and the biopharmaceutics drug disposition classification system. Drug Metab. Dispos. 44, 1731–1735.
Bowman, C.M., Benet, L.Z., 2019. In vitro-in vivo extrapolation and hepatic clearance-dependent underprediction. J. Pharm. Sci. 108, 2500–2504.
Chiba, M., Ishii, Y., Sugiyama, Y., 2009. Prediction of hepatic clearance in human from *in vitro* data for successful drug development. The AAPS J. 11, 262–276.
Dong, J., Park, M.S., 2018. Discussions on the hepatic well-stirred model: re-derivation from the dispersion model and re-analysis of the lidocaine data. Eur. J. Pharm. Sci. 124, 46–60.
Jones, H., Rowland-Yeo, K., 2013. Basic concepts in physiologically based pharmacokinetic modeling in drug discovery and development. CPT 2, e63.
Jones, H.M., Parrott, N., Jorga, K., Lavé, T., 2006. A novel strategy for physiologically based predictions of human pharmacokinetics. Clin. Pharmacokinet. 45, 511–542.
Lombardo, F., Waters, N.J., Argikar, U.A., Dennehy, M.K., Zhan, J., Gunduz, M., Harriman, S.P., Berellini, G., Liric Rajlic, I., Obach, R.S., 2013. Comprehensive assessment of human pharmacokinetic prediction based on *in vivo* animal pharmacokinetic data, part 2: clearance. J. Clin. Pharmacol. 53, 178–191.
Musther, H., Gill, K.L., Chetty, M., Rostami-Hodjegan, A., Rowland, M., Jamei, M., 2015. Are physiologically based pharmacokinetic models reporting the right c max? central venous versus peripheral sampling site. AAPS J. 17, 1268–1279.
Pang, K.S., Han, Y.R., Noh, K., Lee, P.I., Rowland, M., 2019. Commentary: hepatic clearance concepts and misconceptions: why the well-stirred model is still used even though it is not physiologic reality? Biochem. Pharmacol. 169, 113596.
Pang, K.S., Rowland, M., 1977. Hepatic clearance of drugs. II. Experimental evidence for acceptance of the "well-stirred" model over the "parallel tube" model using lidocaine in the perfused rat liverin situ preparation. J. Pharmacokinet. Biopharm. 5, 655–680.
Peters, S.A., 2012. Physiologically-based Pharmacokinetic (PBPK) Modeling and simulations: principles, methods, and Applications in the Pharmaceutical Industry. John Wiley & Sons.
Poulin, P., Kenny, J.R., Hop, C.E., Haddad, S., 2012. In vitro–in vivo extrapolation of clearance: modeling hepatic metabolic clearance of highly bound drugs and comparative assessment with existing calculation methods. J. Pharm. Sci. 101, 838–851.
Rodgers, T., Leahy, D., Rowland, M., 2005. Physiologically based pharmacokinetic modeling 1: predicting the tissue distribution of moderate-to-strong bases. J. Pharm. Sci. 94, 1259–1276.
Watanabe, T., Kusuhara, H., Maeda, K., Shitara, Y., Sugiyama, Y., 2009. Physiologically based pharmacokinetic modeling to predict transporter-mediated clearance and distribution of pravastatin in humans. J. Pharmacol. Exp. Ther. 328, 652–662.

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