Supplemental Data

In Silico Prediction of the Absorption and Disposition of Cefadroxil in Humans using an Intestinal Permeability Method Scaled from Humanized *PepT1* Mice

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Method to Estimate the Concentration-Dependent Permeability of Cefadroxil in Mouse Intestinal Segments during Single Escalating Doses and during Multiple Dosing. The jejunal permeability values of cefadroxil were obtained from wildtype and *huPepT1* mice during *in situ* perfusions of the drug over a concentration range of 0.01-25 mM (Hu and Smith, 2016). The concentration-dependent permeability of cefadroxil (shown in Figure S1) was then fit to the equation:

$$P_{eff} = P_{eff.max}\left(1 - \frac{I}{IC_{50} + I}\right)$$

where $P_{eff.max}$ is the jejunal permeability of cefadroxil at 10 μM, a concentration that is at least 270x lower than the drug's Km value in both genotypes, I is the inhibitor concentration of cefadroxil in this self-inhibition study, and IC_{50} is the concentration of inhibitor at which the P_{eff} of cefadroxil is one-half its maximum value. The parameter estimates are shown in Table S1.

The observed jejunal P_{eff} values of 10 μM cefadroxil in wildtype and *huPepT1* mice are presented in Table S2, along with P_{eff} estimates made in mouse jejunum from human single escalating doses (i.e., 4.1 mM at 5 mg/kg, 12.4 mM at 15 mg/kg, 24.8 mM at 30 mg/kg) and during multiple dosing [i.e., 5.5 mM at 500 mg (6.7 mg/kg) every 6 hours].

The P_{eff} values of 10 μM cefadroxil in other intestinal segments were obtained from wildtype and *huPepT1* mice during *in situ* perfusions of the drug (Hu and Smith, 2016). P_{eff} estimates at other dose levels were then determined by proportionality (Table S2). For example, the duodenal P_{eff} of cefadroxil at 5 mg/kg was estimated in wildtype mice as: $0.40/0.62x0.55=0.36x10^{-4}$ cm/sec. These values in mice were then scaled for human subjects by: $P_{eff.human} = P_{eff.mouse} \bullet (R_{human}/R_{mouse})$ where R is radius of intestine (Tables 3 and S2).

Method to Estimate the Permeability of Cefadroxil in Intestinal Segments of *huPepT1* Mice during the 5 mg/kg Cefadroxil ± 45 mg/kg Cephalexin Drug-Drug Interaction Study. Since the *in situ* intestinal permeability of cephalexin was unknown, we assumed that cefadroxil and cephalexin had the same IC_{50} values. Thus, cephalexin equivalents (i.e., 38.9 mM at 45 mg/kg) were added to cefadroxil (i.e., 4.1 mM at 5 mg/kg) and, at a total concentration of 43.0 mM, the jejunal permeability of cefadroxil in *huPepT1* mice was estimated according to the above equation as $0.023x10^{-4}$ cm/sec. P_{eff} estimates in the other intestinal segments were then determined by proportionality (Table S2). For example, the duodenal P_{eff} in *huPepT1* mice was estimated as: $0.023/0.111x0.154=0.032x10^{-4}$ cm/sec. As before, these values in mice were then scaled for human subjects by: $P_{eff.human} = P_{eff.mouse} \bullet (R_{human}/R_{mouse})$ where R is radius of intestine. (Tables 3 and S2).

TABLE S1

Model fitting of permeability parameters in wildtype (WT) and humanized (hu) PepT1 mice

Parameter	WT	hu
$P_{eff.max}$ (cm/sec x 10^{-4})	0.71 ± 0.07	0.18 ± 0.04
IC_{50} (mM)	4.85 ± 1.73	6.52 ± 4.26

TABLE S2

Observed and predicted intestinal P_{eff} of cefadroxil in wildtype (WT) and humanized (hu) PepT1 mice when estimated by the *in situ* permeability of small and large intestines

	Mouse	Human	**WT**	**hu**	WT	hu	WT	hu	WT	hu	WT	hu	hu
	Radius (cm)		**10 μM**		5 mg/kg		15 mg/kg		30 mg/kg		500 mg (6.7 mg/kg)		CEF + CEP
Stomach	0.40	9.67	**0.00**	**0.00**	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Duodenum	0.14	1.53	**0.55**	**0.24**	0.361	0.154	0.201	0.088	0.128	0.053	0.317	0.136	0.032
Jejenum 1	0.13	1.45	**0.62**	**0.17**	0.402	0.111	0.223	0.063	0.142	0.038	0.352	0.098	0.023
Jejenum 2	0.12	1.29	**0.62**	**0.17**	0.402	0.111	0.223	0.063	0.142	0.038	0.352	0.098	0.023
Ileum 1	0.11	1.13	**0.29**	**0.12**	0.190	0.078	0.106	0.044	0.067	0.027	0.167	0.069	0.016
Ileum 2	0.10	0.98	**0.29**	**0.12**	0.190	0.078	0.106	0.044	0.067	0.027	0.167	0.069	0.016
Ileum 3	0.09	0.82	**0.29**	**0.12**	0.190	0.078	0.106	0.044	0.067	0.027	0.167	0.069	0.016
Caecum	0.62	3.39	**0.00**	**0.06**	0.003	0.036	0.001	0.020	0.001	0.012	0.002	0.032	0.007
Ascending Colon	0.33	2.41	**0.00**	**0.06**	0.003	0.032	0.001	0.018	0.001	0.011	0.002	0.029	0.007

P_{eff}, intestinal permeability in units of cm/sec (x 10^{-4}).

Observed P_{eff} values of 10 μM cefadroxil in the small and large intestines of WT and *huPepT1* mice were obtained previously by our laboratory (Hu and Smith, 2016), and shown in bold text.

Predicted P_{eff} values of cefadroxil in the small and large intestines of WT and *huPepT1* mice are shown after human single oral doses of 5, 15 and 30 mg/kg, after 500 mg (6.7 mg/kg) oral doses every six hours, and after 5 mg/kg cefadroxil plus 45 mg/kg cephalexin (CEF + CEP).

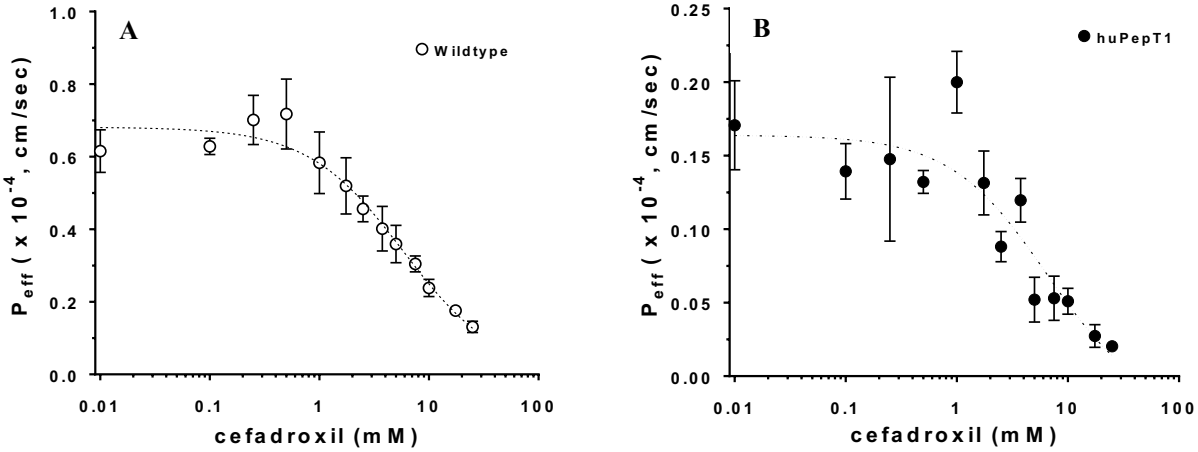

Figure S1. The concentration-dependent jejunal permeability of cefadroxil in wildtype (panel A) and humanized mice (panel B).