

A physiologically based pharmacokinetic model for oxytetracycline residues in sheep

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A physiologically based pharmacokinetic model (PBPK) for oxytetracycline (OTC) residues in sheep was developed using previously published data from a combined serum pharmacokinetic and tissue residue study [Craigmill *et al.* (2000) *J. Vet. Pharmacol. Ther.* 23, 345]. Physiological parameters for organ weights and tissue blood flows were obtained from the literature. The tissue/serum partition coefficients for OTC were estimated from the serum and tissue residue data obtained at slaughter. The model was developed to include all of the tissues for which residue data were available (serum, kidney, liver, fat, muscle and injection site), and all of the remaining tissues were combined into a slowly perfused compartment with low permeability. Total body clearance of OTC calculated in the previous study was used as the starting value for clearance in the PBPK model, with the kidney being the only eliminating organ. The model was built using ACSL (Advanced Continuous Simulation Language) Graphic Modeler®, and the model was fit to the serum and tissue data using the ACSL Math/Optimizer® software (AEGIS Technologies Group, Inc., Huntsville, AL, USA). A sensitivity analysis was also performed to determine which parameters had the greatest effect on the goodness of fit. Numerous strategies were tested to model the injection site, and a model providing a biexponential absorption of the drug from the injection bolus gave the best fit to the experimental data. The model was validated using the clearance parameters calculated from the traditional pharmacokinetic model for each individual animal in the PBPK model. This simple PBPK model well predicted OTC residues in sheep tissues after intramuscular dosing with a long-acting preparation and may find use for other species and other veterinary drugs.

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INTRODUCTION

Physiologically based pharmacokinetic (PBPK) modeling has found extensive use in toxicology and predictive risk assessment for modeling of chemical exposures among various laboratory animals and extrapolation to humans (Charnick *et al.*, 1995; Clewell, 1995; Haber *et al.*, 2001). One of the advantages of PBPK modeling is the ability to predict tissue concentrations of chemicals based on physiological and metabolic parameters for each species of interest. This ability of PBPK modeling to predict tissue concentrations of chemicals at various times after complex exposures has been applied to drugs in food animals in only a few cases. One of the first applications of PBPK to food animals was in salmon to model the disappearance of oxytetracycline (OTC)

from edible tissues (Brocklebank *et al.*, 1997). This model was based on two earlier works that developed PBPK models for exposure to waterborne contaminants (Nichols *et al.*, 1990; Law *et al.*, 1991). An abstract showing a PBPK model developed for OTC in cattle serum and tissues has been presented (Achenbach *et al.*, 1998); however, details have not yet been published. The application of a PBPK model to the prediction of withdrawal times for OTC in Chinook salmon has demonstrated the applicability of PBPK modeling to the prediction of tissue residues in food animals and the establishment of withdrawal periods (Law, 1999). The purpose of the work described in this paper was to develop, apply and validate a PBPK model for the prediction of tissue residues of OTC in sheep after intramuscular (i.m.) dosing with a long-acting formulation.

MATERIALS AND METHODS

Model

The model was developed using Advanced Continuous Simulation Language (ACSL®, Aegis Technologies Group, Inc., Huntsville, AL, USA), which includes a Graphic Modeler® (Version 4.9.4) component for model development and an ACSL Math/Optimizer® (Version 2.5.4) component for fitting the model to the data, and for predictions and sensitivity analysis.

The model was developed to be useful for predictive purposes, so it was constructed in the simplest possible manner that would give a good fit to the experimental data. Therefore, all of the measured tissues were included as compartments (blood, muscle, injection site, liver, kidney and fat) and an additional compartment was used to model all of the other tissues for which time/concentration data were not collected. Physiological and anatomical parameters for adult sheep were obtained from the literature for model development. These parameters and their sources are presented in Tables 1 & 2. After its development, the model was fit to serum and tissue residue data (Craigmill *et al.*, 2000). Individual animal serum data, which were not published, were used in addition to the individual animal tissue data points.

As enterohepatic recirculation was not considered to be necessary for this model, the hepatic blood flow was modeled as a single value with the arterial and portal flows combined. This six-compartment model is shown in Fig. 1. All of the anatomical/physiological parameters were included in an unconnected 'block' (compartment) that contained all the global values for the model. The differential equations that describe each

Table 1. Organ weights of adult sheep as percentage of body weight (MacGregor, 1980; Fluharty *et al.*, 1999)

Tissue weight/volume	As fraction of bodyweight
Blood	0.045
Fat	0.168
Muscle	0.277
Liver	0.015
Kidney	0.003
Injection site*	0.5 kg fixed*
All other tissues	0.362 (based on other tissues and GI contents)
GI contents†	0.13

*Included in muscle fraction. † Not included in the model.

Table 2. Tissue blood flow as a percentage of cardiac output

Tissue blood flow	Fraction of cardiac output	References
Cardiac output	6.9 L/h/kg (115 mL/min/kg)	Talke <i>et al.</i> (1985), Evans <i>et al.</i> (1998), Dodic <i>et al.</i> (2001), Ullman <i>et al.</i> (2001)
Fat	0.06–0.15	Gregory <i>et al.</i> (1986), Hales & Fawcett (1993)
Muscle (and injection site)	0.224	Talke <i>et al.</i> (2000), Zheng <i>et al.</i> (2000)
Liver	0.183 (includes arterial and portal flow)	Kisauzi & Leek (1991), Schiffer <i>et al.</i> (1993)
Kidney	0.064	Tsuji <i>et al.</i> (1985), Talke <i>et al.</i> (2000), Ullman <i>et al.</i> (2001)
Other tissues	0.473	Calculated from difference

compartment are presented in Table 3. Details of the ACSL® coding and equations for individual compartments are presented in Appendix 1. As OTC is excreted primarily in urine, and total body clearance of OTC and renal clearance are virtually identical in cattle (Nouws *et al.*, 1985), the kidney was the only organ that included a clearance parameter. The starting clearance value for model fitting was the average clearance (1.74 mL/min kg) found for sheep in an earlier study (Craigmill *et al.*, 2000).

The injection bolus was not considered as a physiological compartment in the PBPK model, but as a separate absorption compartment without blood flow. The input from the injection bolus was into the injection site muscle, not directly into the blood compartment. Absorption from the injection bolus into the 500-g muscle injection site was modeled using a biexponential process, in which the slower phase rate constant and fraction of the dose allocated to the slower phase were dependent on the fast phase rate constant and fraction of the dose allocated to the fast phase. These parameters were purposefully made very flexible to find the optimum values for fitting the model. The equation describing the absorption process is

$$Amount = (Dose \times Fx) \times e^{-k_{fast} \times t} + [Dose \times (1 - Fx)] \times e^{-k_{slow} \times t}$$

where *Amount* is the amount remaining at the injection site, *Fx* is the fraction of the dose allocated to the fast phase of absorption governed by the fast rate constant of absorption k_{fast} , *Dose* is the total dose delivered, and k_{slow} is the slow rate constant of absorption defined as fraction of k_{fast} .

Cardiac output values were adjusted to reflect organ plasma perfusion by including a parameter for the hematocrit. The hematocrit values were obtained from an earlier unpublished study involving 20 normal, healthy sheep (10 wethers and 10 females). The mean value, 0.333, of all the animals was used.

The injection sites collected previously (Craigmill *et al.*, 2000) consisted of 0.5 kg of muscle tissue surrounding the i.m. injection site. The injection site was modeled as a separate 0.5-kg 'sub-compartment' of the total body muscle. The 'baseline' concentration of OTC in the 0.5-kg injection site was the same as that calculated for total body muscle using the muscle partition coefficient ($R_{muscle} = R_{IS}$) and fractional blood flow to the injection site (Q_{IS}). Thus, the concentration of OTC in the injection site muscle was the sum of the 'baseline' concentration in the 0.5 kg of muscle, plus the remaining amount of unabsorbed drug in the injection 'bolus' divided by 0.5 kg.

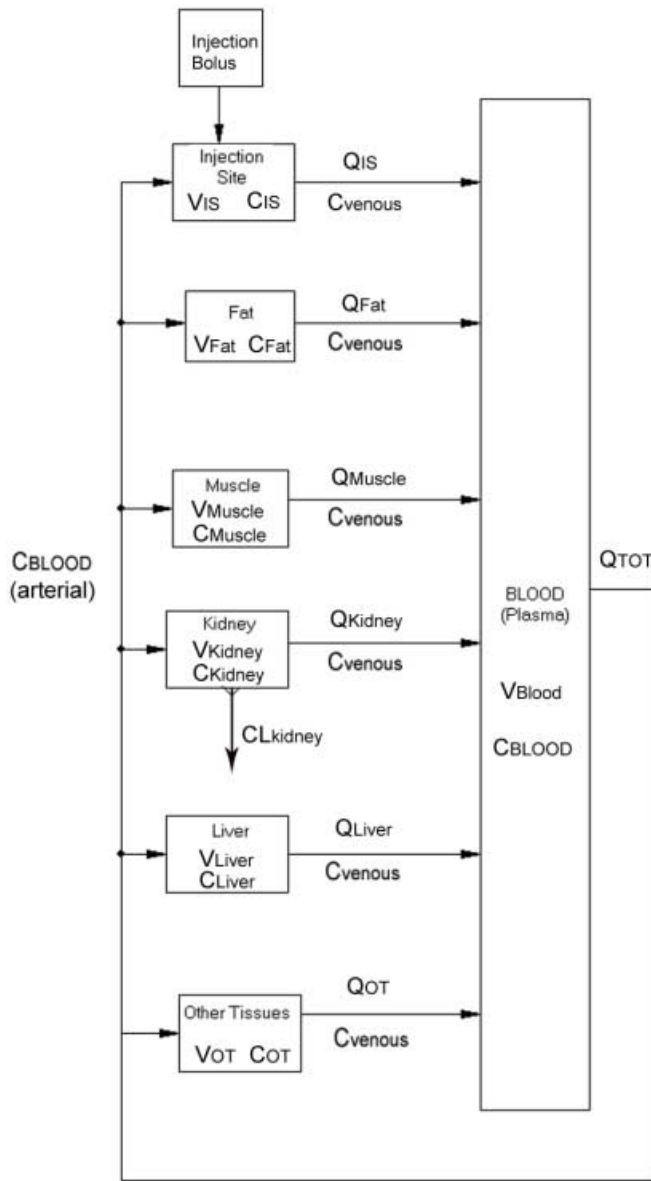


Fig. 1. PBPK model for oxytetracycline in sheep. *V*, tissue volumes; *C*, tissue concentrations; *Q*, tissue blood (plasma) flow; *CL_{kidney}* renal clearance; *C_{venous}* venous blood concentration which equals the tissue concentration divided by the tissue partition coefficient; *Q_{TOT}* cardiac output.

Table 3. Equations describing the rate of change of oxytetracycline concentration in each tissue

Muscle	$\frac{dC_{\text{muscle}}}{dt} = \frac{[C_{\text{blood}} - (C_{\text{muscle}}/R_{\text{muscle}})] \cdot Q_{\text{muscle}}}{V_{\text{muscle}}}$
Injection site	$\frac{dC_{\text{IS}}}{dt} = \frac{[C_{\text{blood}} - (C_{\text{IS}}/R_{\text{IS}})] \cdot Q_{\text{IS}} + [(k_{\text{slow}} \cdot (1-F) \cdot BW \cdot DOSE) + (k_{\text{fast}} \cdot F \cdot BW \cdot DOSE)]}{0.5}$
Liver	$\frac{dC_{\text{liver}}}{dt} = \frac{[C_{\text{blood}} - (C_{\text{liver}}/R_{\text{liver}})] \cdot Q_{\text{liver}}}{V_{\text{liver}}}$
Fat	$\frac{dC_{\text{fat}}}{dt} = \frac{[C_{\text{blood}} - (C_{\text{fat}}/R_{\text{fat}})] \cdot Q_{\text{fat}}}{V_{\text{fat}}}$
Other tissues	$\frac{dC_{\text{OT}}}{dt} = \frac{[C_{\text{blood}} - (C_{\text{OT}}/R_{\text{OT}})] \cdot Q_{\text{OT}}}{V_{\text{OT}}}$
Kidney	$\frac{dC_{\text{kidney}}}{dt} = \frac{[(C_{\text{blood}} - (C_{\text{kidney}}/R_{\text{kidney}})) \cdot Q_{\text{kidney}} - C_{\text{kidney}} \cdot CL_{\text{kidney}}]}{V_{\text{kidney}}}$
Blood (plasma)	$\frac{dC_{\text{blood}}}{dt} = \frac{\{[(C_{\text{liver}}/R_{\text{liver}}) \cdot Q_{\text{liver}}] + [(C_{\text{kidney}}/R_{\text{kidney}}) \cdot Q_{\text{kidney}}] + [(C_{\text{muscle}}/R_{\text{muscle}}) \cdot Q_{\text{muscle}}] + [(C_{\text{fat}}/R_{\text{fat}}) \cdot Q_{\text{fat}}] + [(C_{\text{OT}}/R_{\text{OT}}) \cdot Q_{\text{OT}}] - C_{\text{blood}} \cdot Q_{\text{blood}}\}}{V_{\text{blood}}}$

C_x = concentration of chemical in each compartment (μg/kg) (ppb). *R_x* = partition coefficient of drug between the tissue and blood plasma. *Q_x* = blood flow to the tissue (L/h). *V_x* = tissue volume (weight) (kg). Subscript *x* = name of the compartment: blood (plasma), fat, muscle, kidney, liver, OT (other tissues), IS (injection site). *k_{slow}* = slow absorption phase rate constant (h⁻¹). *k_{fast}* = rapid phase absorption rate constant (h⁻¹). *F* = fraction of dose allocated to fast absorption phase. *BW* = body weight (kg). *DOSE* = dose (μg per kg body weight).

Model fitting procedures

The ACSL software contains an optimization routine for fitting models to experimentally collected data. Model parameters can be adjusted to achieve the best possible fit to the experimental data points using a maximum likelihood estimation algorithm (Generalized Reduced Gradient included in the ACSL Math/Optimizer®). Limits are set on how much parameters can be adjusted to insure that the results are biologically plausible. After extensive fitting runs to test the effects of modifying various parameters on the ‘fit’, the optimization procedure was limited to nine parameters. For this study, the parameters that were included in the optimization procedure were the fast rate constant of absorption, the percentage of dose handled by the fast phase, the slower rate constant of absorption (as a fraction of the fast constant), the total body clearance, the partition coefficients of the muscle, fat, liver and kidney tissues, and the blood flow to fat. Starting values and limits for these parameters are shown in Table 4.

Model validation

In order to validate the usefulness of the model for predicting tissue residues, the clearance parameters previously calculated from the serum pharmacokinetics for individual sheep were used to predict tissue concentrations at their time of slaughter using the PBPK model. All of the PBPK model parameters were set to those that were optimal for fitting the model to all of the animal data. The clearance parameter in the PBPK model was then set to the value derived from the serum data for each individual animal, and the model run to predict all of the tissues at the time of slaughter. The observed and predicted values were plotted and linear regressions performed for each tissue.

RESULTS

The final (optimal) parameters obtained from the optimization of the model to the experimental data are presented in Table 4. The optimized parameters are very similar to the initial starting values for the parameters except for the kidney partition coefficient (*P*), which was increased by approximately 30% over

Parameter	Lower bound	Starting value	Upper bound	Final, mean \pm SD
Fast absorption K_{fast}	0.01	0.032	0.1	0.0427 \pm 0.0039
Fast absorption fraction	0.5	0.714	0.99	0.704 \pm 0.083
Slow absorption fraction of K_{fast}	0.1	0.25	0.5	0.298 \pm 0.053
Kidney clearance (mL/min kg)	1	1.74	4	2.49 \pm 0.09
Kidney P	3	4.75	8	6.46 \pm 0.44
Liver P	1	1.89	3	1.82 \pm 0.13
Fat P	0.02	0.086	0.1	0.0853 \pm 0.0063
Muscle P	0.5	0.85	1.2	0.878 \pm 0.045
Fat Q (%CO)	0.06	0.09	0.15	0.06 \pm 0.813

Table 4. Model fitting parameter limits and final values. Starting P (serum/tissue partition coefficient) values obtained from Table 3, Craigmill *et al.*, 2000

the observed P from the terminal phase of the tissue residue study, and the clearance parameter that was increased approximately 40%. Plots of the resulting fit of the model to the residue data are shown in Fig. 2. Plots of the standardized residuals for each tissue shown in Fig. 3 show even distributions about zero for all tissues at all times except for the liver, in which the residuals for the 24-h time-point were all positive. Sensitivity analysis of these parameters showed that the fast rate constant of absorption and the renal clearance were the most important parameters for determining the OTC residue concentration in each tissue over time.

The results of the validation procedure are shown in Fig. 4. The regression results for comparing the model predicted and the observed tissue concentrations are presented in Table 5. The injection site residues were the only ones that were not well predicted by the model. In all cases except the injection site, the model over predicts the residues at each time; however, the over prediction is consistent for each tissue and can be accounted for

using the equation for each tissue plot of observed vs. predicted residue.

DISCUSSION

Several papers have been published using PBPK modeling to predict tissue residues of drugs in fish (Brocklebank *et al.*, 1997; Law *et al.*, 1991; Law, 1999) and one abstract has been presented on work done to develop a PBPK model for OTC in cattle blood and tissues (Achenbach *et al.*, 1998). Unfortunately, a full publication of this model has not been published; however, the abstract describes work in cattle that is virtually identical to what has been done in this paper for sheep. This paper presents the first use of PBPK modeling for tissue residues and serum pharmacokinetics of a drug in sheep. The validation of the model using the clearance parameters calculated from only serum data showed that the model works well. The consistent over

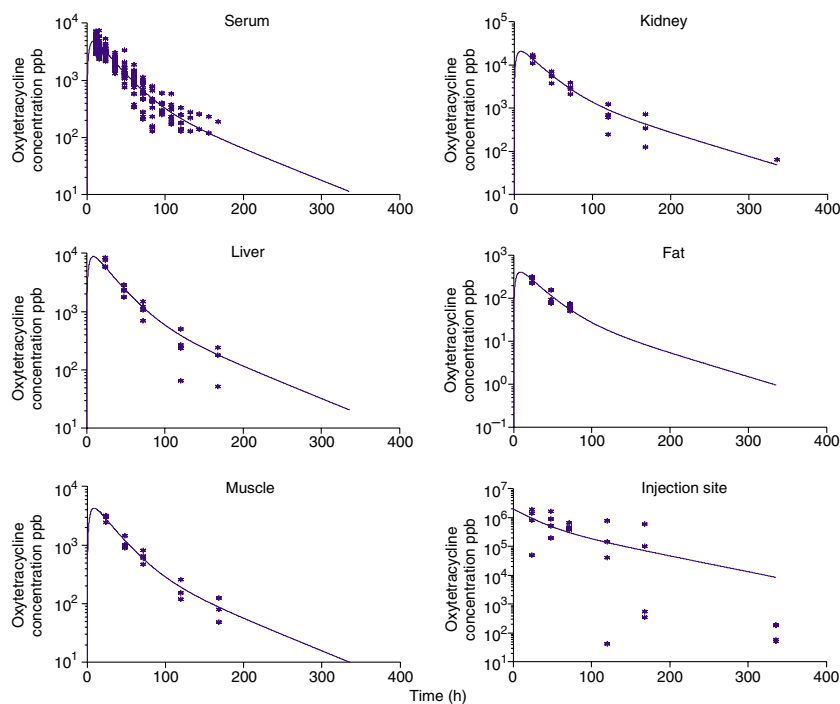


Fig. 2. Final fits to tissue residue data with predicted lines and observed data points.

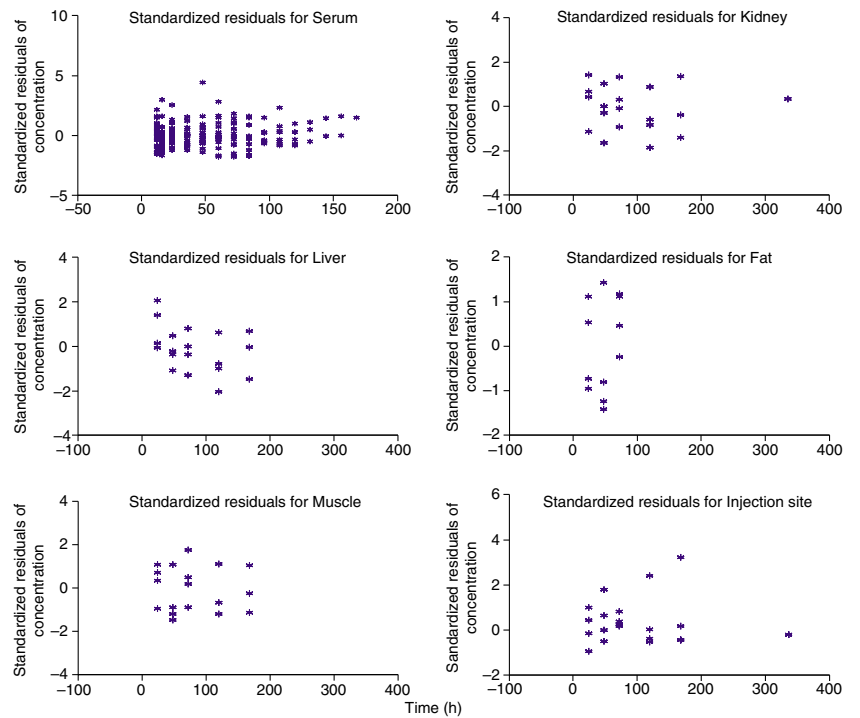


Fig. 3. Standardized residual plots for line fits of each tissue.

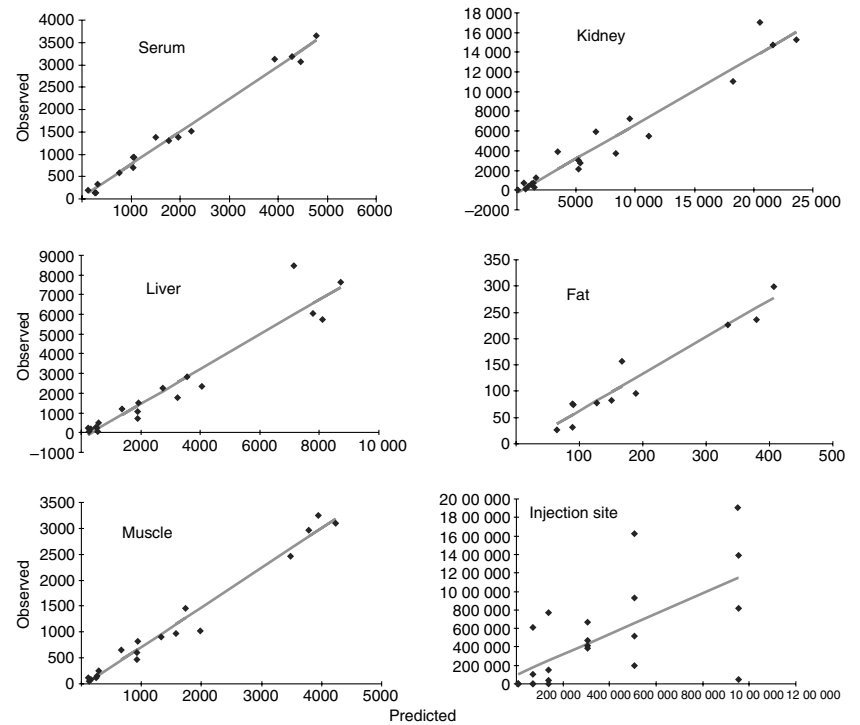


Fig. 4. Model validation plots of observed data and predicted data using individual animal clearance parameters and earlier serum pharmacokinetic study (Craigmill *et al.*, 2000).

prediction of all the tissues except the injection site when using the individual animal clearance values may be because of incomplete absorption of OTC from the injection site. The starting value for clearance of 1.74 is 70% of the optimal value

determined during the fitting procedure (2.48). The slope values for fitting the observed vs. predicted data range from 0.69 to 0.88, with an average of 0.75 for all the tissues excluding the injection site. This suggests that the fraction of drug absorbed

Table 5. Model validation results comparing the model predicted tissue concentrations at slaughter to those observed at slaughter, using the optimal model parameters and the unpublished clearance parameters for individual animals from an earlier study (Craigmill *et al.*, 2000)

Tissue	Slope (<i>m</i>)	Intercept (<i>b</i>)	R ²
Serum	0.733 ± 0.022	42.2	0.987
Kidney	0.690 ± 0.037	-248	0.950
Liver	0.88 ± 0.06	-301	0.923
Fat	0.70 ± 0.06	-7.1	0.928
Muscle	0.76 ± 0.03	-50.4	0.977
Injection site	1.10 ± 0.27	97, 720	0.432

may be less than 1 and is likely to be approximately 75%. This would result in an overestimation of the concentrations with 100% bioavailability assumed in the model.

Optimally, an external data set would be used to validate that this model is predictive outside of the data set used to develop the model. Another data set of OTC tissue residues in sheep is not available to do this, thus external validation is not possible at this time.

Oxytetracycline is a relatively simple drug to work with for PBPK modeling because it is not metabolized and is excreted unchanged in urine. In addition, its distribution to edible tissues appears to be governed primarily by its partition coefficient in those tissues. It is interesting to note that the partition coefficient for OTC into fat as calculated by this model is 0.086. Three values listed for the log *P* of OTC in octanol/water media are -0.96, -1.12, and -1.6 (Riviere *et al.*, 1991). When converted to fractions, these values are 0.11, 0.076, and 0.025, respectively (mean = 0.070), which corresponds closely with the model optimized *P* value for OTC in sheep fat/blood (0.085). This suggests that for some drugs it may be possible to use octanol/water partition coefficients as starting approximations for tissue partition coefficients if blood/tissue data are unavailable.

This simple model can serve as a starting point for further exploration of the use of PBPK models in the prediction of drug residues in ruminants. Further studies are planned to apply this model to data in cattle and goats, and for other drugs.

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APPENDIX 1

Block (compartmental) parameters for ACSL® model

The following information is taken directly from the blocks that were used in the PBPK Sheep model. The blocks are modifications of standard blocks included with the ACSL® Tox toolkit using the ACSL Graphic Modeler module. The blocks were originally written by Xiaofeng Wang of MGA Inc. in 1996 and are currently copyrighted by Aegis Technologies Group, 2001, as part of the ACSL Simulation Software, Version 11.8.

Global parameter block

constant BODYWEIGHT = 50	! Animal weight in kilograms
constant Qcar = 6.9	! units are in liters per hour per kilogram body weight
constant WDOSE = 20000	! dose in µg/kg
constant QcM = 0.224	! Muscle blood flow fraction of cardiac output
constant QcFAT = 0.091	! Fat blood flow fraction of cardiac output
constant QcKID = 0.064	! Kidney blood flow fraction of cardiac output
constant QcLIVER = 0.183	! Liver blood flow fraction of cardiac output
constant VINJEXSITE = 0.5	! Injection site volume = 0.5 kg, not a fraction of Body weight
constant VFATFX = 0.168	! Fat volume, fraction of body weight
constant VMUSCLEFX = 0.277	! Muscle volume, fraction of body weight
constant VLIVERFX = 0.015	! Liver volume, fraction of body weight
constant VKIDNEYFX = 0.003	! Kidney volume, fraction of body weight
constant VBLOODFX = 0.045	! Blood volume, fraction of body weight
constant HEMATOCRIT = 0.333	! hematocrit (PCV)
constant GICONTENTFX = 0.13	! fraction of BW which is GI contents
VGICONTENTS = GICONTENTFX * BODYWEIGHT	! Volume (weight) of GI contents
VFAT = VFATFX * BODYWEIGHT	! Fat Volume
VMUSCLE = (VMUSCLEFX * BODYWEIGHT)	! Muscle volume, includes the injection site
VLIVER = VLIVERFX * BODYWEIGHT	! Liver Volume
VKIDNEY = VKIDNEYFX * BODYWEIGHT	! Kidney Volume
VBLOOD = (VBLOODFX * BODYWEIGHT) * (1-HEMATOCRIT)	! Serum Volume calculation
VOT = BODYWEIGHT - (VFAT + VMUSCLE + VLIVER + VKIDNEY + VBLOOD + VGICONTENTS)	! Volume other tissues determined by difference
QTOT = (Qcar * BODYWEIGHT) * (1 - HEMATOCRIT)	! Cardiac Output of serum
QcIS = QcM * (VINJEXSITE/VMUSCLE)	! Injection site blood flow fraction
QcOT = 1 - (QcM + QcFAT + QcKID + QcLIVER)	! Other tissue blood flow fraction

IM/SC absorption block (biexponential absorption process)

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constant F = 0.714                ! fraction of dose allotted to rapid phase of absorption
a0 = F * BODYWEIGHT * WDOSE      ! Starting amount of drug in bolus at Injection site at time zero, allocated to rapid phase absorption
amnt = a0
a1 = (1 - F) * (BODYWEIGHT * WDOSE) ! Starting amount of drug in bolus at Injection site at time zero, allocated to slow phase absorption
amnt2 = a1
constant k = 0.032334            ! Absorption rate constant to injection site compartment (L/time)
constant KK = 0.25               ! Fraction of constant k which determines the slow rate constant of absorption, k1
k1 = k * KK                      ! Slow rate constant of absorption
ro = k * amnt                    ! Rate for fast phase (mass/time)
r1 = k1 * amnt2                  ! Rate for slow phase (mass/time)
rate = -ro
rate2 = -r1
rate3 = (ro + r1)                ! Sum of rates for input into Injection site compartment
amnt = integ(rate, a0)           ! ACSL code for integration of fast phase
amnt2 = integ(rate2, a1)         ! ACSL code for integration of slow phase
DOSEMASS = amnt + amnt2          ! Total amount remaining at the site of absorption for calculating injection site concentration

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Injection site block

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V = VINJEXSITE                    ! Injection site volume (0.5 kg fixed)
Q = QcIS * QTOT                  ! Blood flow rate through the tissue (volume/time)
constant P = 0.841               ! Tissue/blood equilibrium distribution ratio
C = (a/(V))                      ! Concentration of chemical in the compartment (mass/volume) excluding the injection bolus
Ctb = C/p                        ! Concentration of chemical in tissue blood (mass/volume)
INJEXSITE = (DOSEMASS/V) + C     ! Injection site concentration equals the amount of unabsorbed drug (DOSEMASS/C) plus muscle concentration
                                ! contributed by blood flow after absorption (C)
r = Q * (Ca - Ctb) + inp         ! Rate of change of amount of chemical in the injection site (mass/time). Note: inp = input rate3 from
                                ! IM/SC Block
a = intvc(r, a0)                 ! ACSL coding for integration of mass balance equation above to calculate amount of chemical in the
                                ! compartment (mass)

```

Fat compartment/block

```

V = VFAT                          ! Fat volume
Q = QcFAT * QTOT                  ! Blood flow rate through the fat (volume/time)
constant P = 0.086                ! Fat/blood equilibrium distribution ratio
constant a0 = 0.0                 ! Initial amount of chemical in the fat
C = a/(V)                         ! Concentration of chemical in the compartment (mass/volume)
Ctb = C/p                         ! Concentration of chemical in tissue blood (mass/volume)
r = Q * (Ca - Ctb)                ! Rate of change of amount of chemical in the fat (mass/time)
a = intvc(r, a0)                  ! ACSL coding for integration of mass balance equation above to calculate amount of chemical in the compartment (mass)

```

Muscle compartment/block

```

V = VMUSCLE                       ! Muscle volume minus injection site 0.5 kg
Q = (QcM - QcIS) * QTOT          ! Blood flow rate through the tissue (volume/time)
constant P = 0.85097              ! Tissue/blood equilibrium distribution ratio
constant a0 = 0.0                 ! Initial amount of chemical in the tissue
C = a/(V)                         ! Concentration of chemical in the compartment (mass/volume)
Ctb = C/p                         ! Concentration of chemical in tissue blood (mass/volume)
r = Q*(Ca - Ctb)                 ! Rate of change of amount of chemical in the muscle (mass/time)
a = intvc(r, a0)                  ! ACSL coding for integration of mass balance equation above to calculate amount of chemical in the compartment
                                ! (mass)

```

Kidney compartment/block

V = VKIDNEY	! Kidney volume
Q = QcKID * QTOT	! Blood flow rate through the tissue (volume/time)
constant P = 4.7458	! Tissue/blood equilibrium distribution ratio
constant a0 = 0.0	! Initial amount of chemical in the tissue
constant Cl = 2.0555	! Total Body clearance in (mL/min)/kg from WinNonlin analysis
C = a/(V)	! Concentration of chemical in the compartment (mass/volume)
Ctb = C/p	! Concentration of chemical in tissue blood (mass/volume)
kex = (Cl/1000) * 60 * BODYWEIGHT	! Clearance (mL/min/kg) based on BW and converted to L/h
rex = kex * C/P	! Rate of excretion (mass/time)
r = Q * (Ca - Ctb) - rex	! Rate of change of amount of chemical in the kidney (mass/time)
a = intvc(r, a0)	! ACSL coding for integration of mass balance equation above to calculate amount of chemical in the compartment (mass)

Liver compartment/block

V = VLIVER	! Liver volume
Q = QcLIVER * QTOT	! Blood flow rate through the tissue (volume/time)
constant P = 1.8967	! Tissue/blood equilibrium distribution ratio
constant a0 = 0.0	! Initial amount of chemical in the tissue
C = a/(V)	! Concentration of chemical in the compartment (mass/volume)
Ctb = C/p	! Concentration of chemical in tissue blood (mass/volume)
r = Q * (Ca - Ctb)	! Rate of change of amount of chemical in the liver (mass/time)
a = intvc(r, a0)	! ACSL coding for integration of mass balance equation above to calculate amount of chemical in the compartment (mass)

Other tissue compartment/block

V = VOT	! Other tissue volume
Q = QcOT * QTOT	! Blood flow rate through the tissue (volume/time)
constant P = 0.1	! Tissue/blood equilibrium distribution ratio
constant a0 = 0.0	! Initial amount of chemical in the tissue
C = a/(V)	! Concentration of chemical in the compartment (mass/volume)
Ctb = C/p	! Concentration of chemical in tissue blood (mass/volume)
r = Q*(Ca - Ctb)	! Rate of change of amount of chemical in the other tissue compartment (mass/time)
a = intvc(r, a0)	! ACSL coding for integration of mass balance equation above to calculate amount of chemical in the compartment (mass)

Blood (plasma) compartment/block

V = VBLOOD	! Plasma compartment volume
constant a0 = 0.0	! Initial amount in venous compartment
	<i>! The coding below in italics sets the maximum and minimum of blood flow rate to insure it is within reasonable bounds. If the flows are outside the bounds set, the model will compile but does not produce numeric results. The bounds must be made wider when investigating blood flow effects on model fitting, or the coding for the flow checking disabled.</i>
constant Qmin = 0.95	
constant Qmax = 1.05	<i>! - - - check if the sum of blood flow rates = QTOT</i>
QCHECK = (Q1 + Q2 + Q3 + Q4 + Q5 + Q6)/QTOT	
termt(QCHECK.lt.Qmin, 'sum of blood flow is less than QTOT')	
termt(QCHECK.gt.Qmax, 'sum of blood flow is larger than QTOT')	
Cv = a/V	Cv = a/V !Concentration of chemical in the compartment (mass/volume)
R = Q1 * Ctb1 + Q2 * Ctb2 + Q3 * Ctb3 + Q4 * Ctb4 + Q5 * Ctb5 + Q6 * Ctb6 - QTOT * Cv	
	! Rate of change of amount of chemical in the blood compartment (mass/time)
a = intvc(r, a0)	! ACSL coding for integration of mass balance equation above to calculate amount of chemical in the compartment (mass)

Copies of this ACSL® model and the full data sets used in this paper are available from the author by request.