MMID4 Manual

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for Mass Transport and Exchange

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MMID4 Manual

Contents

1. MMID4 OVERVIEW

1.1. What Is MMID4?

The name, MMID4, is an acronym for Multiple path, Multiple tracer, Indicator Dilution, 4 region model. The MMID4 model is a simulation of blood–tissue exchange that can be used to examine the behavior of three types of tracers: a type of tracer that remains in the vasculature, a type that can leave the vasculature but that remains extracellular, and a fully permeant tracer that enters cells. MMID4 can be used to study the physiology of the exchange process, or as a tool to help analyze experimental data. The exchange process may be examined by viewing the instantaneous outflow concentration, the instantaneous extraction, or the amount of tracer remaining in the exchange system (i.e., the residue function).

The model provides ways to examine tracer delay and dispersion between the injection site and the target organ, and the effects of flow heterogeneity on exchange within the organ. Fig. 1.1 shows the general layout of the model. Each of the multiple tracers flows through the common arterial nonexchanging vessels, through the parallel pathways each containing an arteriole, exchange unit, and venule, and then through the common venous nonexchanging vessels.

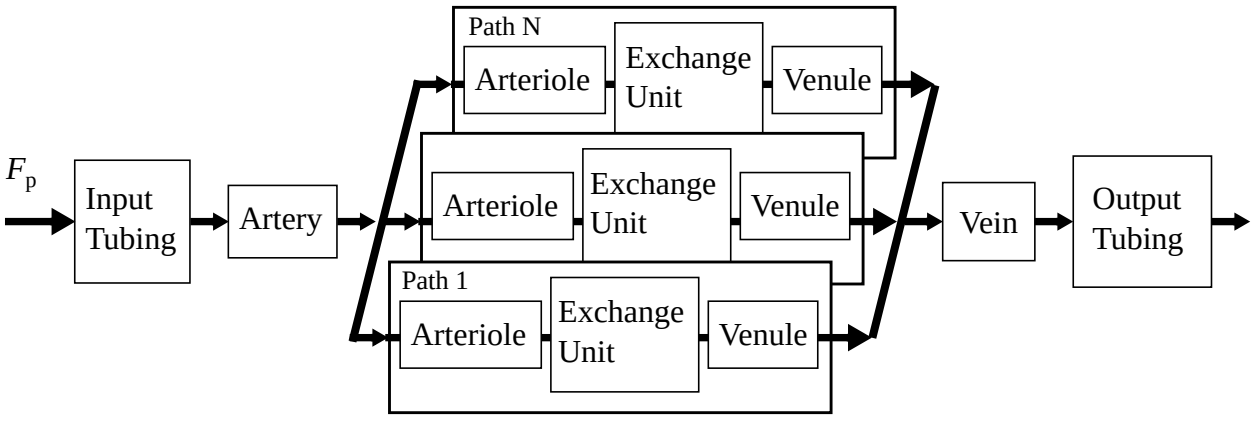


Figure 1.1. MMID4 provides up to 20 pathways to account for tissue heterogeneity. Flow enters each pathway after passing through optional large-vessel components. Each pathway includes optional small-vessel components and one exchange unit. Large-vessel and small-vessel components account for delay and dispersion of the input function before it reaches the exchange unit.

In formal notation, the outflow concentration-time curve seen from an exchange unit in response to an impulse input of tracer is denoted as $h(t)$. We use $h(t)$ as a general symbol to denote the normalized (unity area) outflow dilution curve even when the input has forms other than an impulse. A subscript is added to designate specific tracers. Thus, $h_D(t)$ denotes the "permeating" or "diffusible" tracer, $h_E(t)$ denotes the extracellular tracer, and $h_R(t)$ denotes the intravascular "reference" tracer.

1.2. Vascular Components

1.2.1. *Nonexchanging vessels*

The nonexchanging vessel operators

The nonexchanging vessels in MMID4 are modeled by vascular operators (King et al., 1993). Each nonexchanging vessel is completely described by its volume and relative dispersion (*RD*). If a particular operator is not desired in a given simulation, it can be effectively removed by setting its volume to zero. Although the nonexchanging vessels are normally used to describe vascular delay and dispersion, they can also describe pure delay (no dispersion) by setting *RD* to zero.

Arterial vessels

Three types of arterial vessels are shown in Fig. 1.1. The first is the input tubing. This models a cannula or catheter through which tracers are injected. While not strictly an arterial vessel, it is upstream of the organ and can be described in the same way as the nonexchanging vessels, by its volume and *RD*.

The second type of arterial vessel, downstream from the input tubing, is the artery. Tracer flowing out of the artery is distributed to the various flow paths where it enters the third type of arterial vessel, the arterioles, and then the exchange units. The distribution of flow among the pathways is controlled by the flow heterogeneity parameters described in Section 3.3.2.

Venous vessels

Fig. 1.1 shows that the venous vessels in MMID4 are similar to the arterial vessels. Each exchange unit drains into a venule. All venules drain into the common vein, then into the output tubing.

1.2.2. *Exchange units*

The blood–tissue exchange unit (BTEX) operator

The basic tissue unit in which exchange takes place, shown in Fig. 1.2, consists of four regions: an intravascular space, the endothelium, the interstitium, and the parenchymal cells. Note that the interstitium (*ISF*) is split into two parts. The first, ISF_1, lies between the endothelial and parenchymal cells. The second, ISF_2, is connected to ISF_1 and the parenchymal cell. This is considered a four region model despite the split of the *ISF* into two parts.

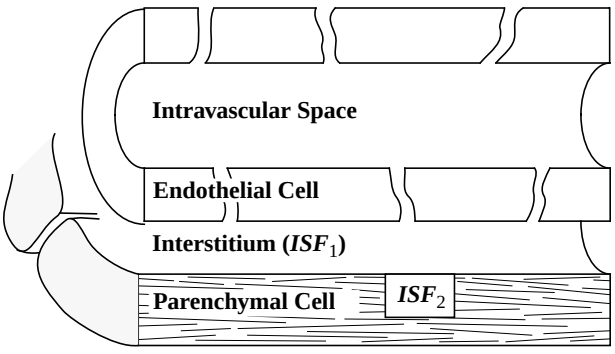


Figure 1.2. Basic exchange unit in the MMID4 model, with four regions in which exchange can take place.

Flow and exchange within the capillary is modeled by a Lagrangian sliding fluid element algorithm, as shown in Fig. 1.3. The exchange unit is divided into a series of axially distributed segments. In the first time step, exchange between the capillary and surrounding tissue is calculated. At the beginning of each subsequent time step, the fluid in the capillary is advanced one segment, and exchange is allowed to occur again. The rationale for this approach has been discussed by Bassingthwaighte (1974) and Bassingthwaighte et al. (1989). The model provides for up to 60 axial segments.

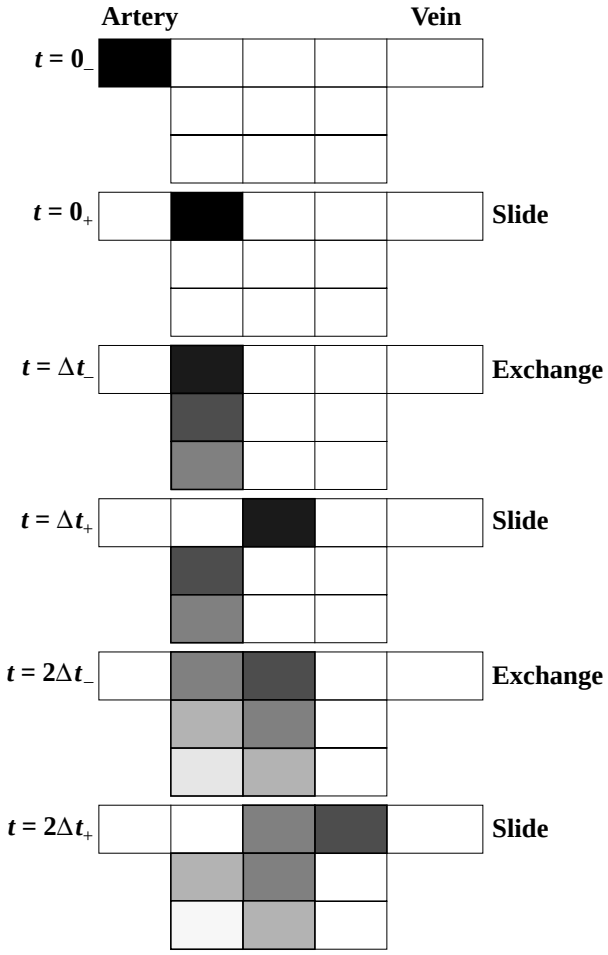


Figure 1.3. Lagrangian sliding element algorithm for blood–tissue exchange: $\Delta t = V_p/(F_p \cdot$ Number of segments). With each Δt, the capillary contents slide instantaneously one segment downstream, after which the radial transmembrane and axial diffusional exchanges, and the consumption, are computed.

Intravascular tracer exchange units

The parameters affecting the intravascular tracer are shown in Fig. 1.4. The volume of distribution within the intravascular region is the plasma volume (V_p). The tracer can undergo axial diffusion (D) and may also be bound to receptors on the endothelial surface. Such binding, if reversible, may be simulated by allowing movement into the interstitium similar to the movement of the extracellular tracer. Hence, PS_g and V'_{isf} are also included as parameters for

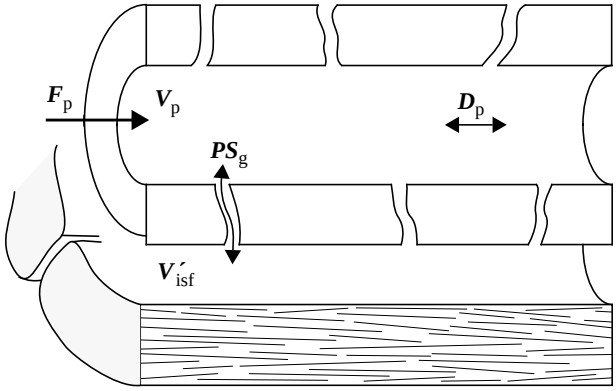


Figure 1.4. Parameters affecting the intravascular tracer. In this case, the *PS* product of the gap (PS_g) and apparent volume of distribution in the interstitial space (V'_{isf}) are included to simulate receptor binding on the endothelial surface. See text for details.

the intravascular tracer. The plasma flow, F_p, and plasma volume, V_p, are the same parameter for all three tracers.

Extracellular tracer exchange units

The parameters affecting the extracellular tracer concentrations are shown in Fig. 1.5. The extracellular tracer has an apparent volume of distribution in the interstitial space (V'_{isf}) and in the plasma (V_p). It can undergo axial diffusion in the intravascular and interstitial regions (D). It can only move between the two regions through the interendothelial cell clefts. Hence, its movement depends on only one *PS* product.

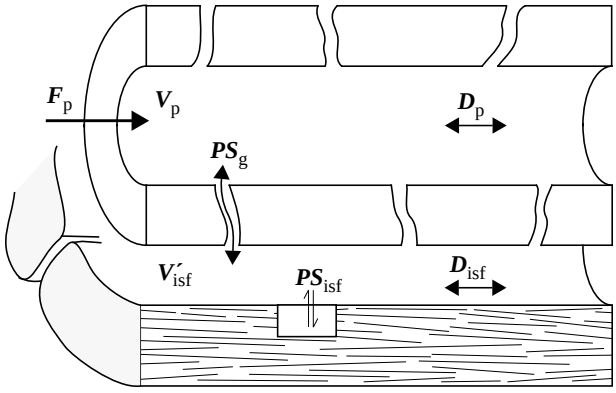


Figure 1.5. Parameters affecting exchange of the extracellular tracer. See text for details.

Permeant tracer exchange units

The parameters affecting the concentration of the permeant tracer in each region are shown in Fig. 1.6. Because this tracer can move to all regions, it has an apparent volume of distribution (V) in all regions. In the intravascular region, this is the plasma volume (V_p). Also in each region, the tracer has the potential to undergo axial diffusion (D). The tracer may be metabolized or undergo chemical reactions that result in its being cleared from the region. The degree to which this happens is indicated by the intraregion consumption or "gulosity" (G). Movement of the tracer between regions is governed by the permeability-surface area product (PS) of the various barriers. A molecule of

tracer may move, for example, between the capillary and the interstitium. Two routes are available; it can either move through the clefts between the endothelial cells, or it can move through the endothelial cell. In the first case, movement depends on one *PS* product (PS_g). In the second, movement depends on the *PS* product at the luminal side of the cell (PS_{ecl}) and the *PS* product on the abluminal side of the cell (PS_{eca}).

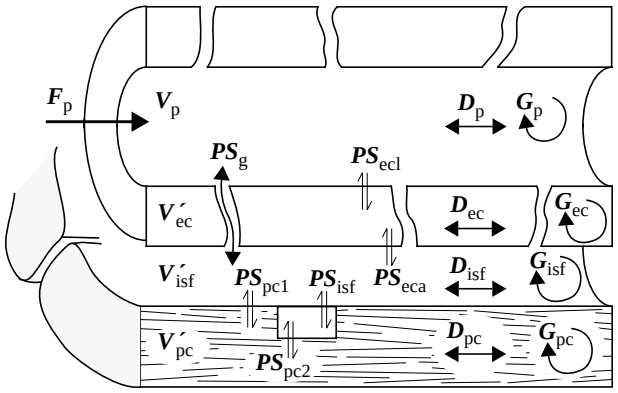


Figure 1.6. Parameters affecting exchange of the permeant tracer. F_p and V_p are the same for all tracers. See text for details.

1.3. Additional Features

1.3.1. *Input function generation*

General MMID4 permits the user to generate an input function in a number of ways. These are discussed briefly below. For a given simulation run, the user selects one of the available methods. Generation of the input function is discussed in detail in Section 3.2 of this manual and is the subject of exercises in Section 2.2.2.

Function generator MMID4 gives the user access to a generalized function generator. It will generate a variety of functions, including several types of pulses and pulse trains and a number of density functions. This generator is named *cinput*: additional information about it can be found in the UNIX online manual page.

Curves from reference data If the user specifies that reference data are to be loaded by XSIM, curves from these data can be used as the input function for one or more of the tracers.

Deconvolution When there are reference data that contain an outflow curve for an intravascular tracer, the user can direct MMID4 to deconvolve those data with the model transfer function and use the result as the input function.

1.3.2. *Heterogeneity*

General When the user configures MMID4 to use more than one flow path, the flow paths may have different flows, membrane conductances, and tracer consumptions. These heterogeneities are used to simulate tracer transport and exchange

in a whole organ. Heterogeneity is discussed in Section 3.3 of this manual and is the subject of exercises in Section 2.4.3.

Flow — In a multipath configuration of MMID4, flows for each path must be specified. These flows can be specified by using a probability density function, by a curve in the reference data, or manually, by the user entering the flow for each path.

Membrane conductance — When multiple paths are specified, the user can specify that the conductances of the capillary wall and endothelial cell for each path be constant or proportional to the flow in that path.

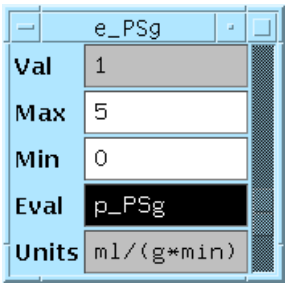
Tracer consumption — As with membrane conductances, the user can specify that the consumption of tracer in the endothelial cells and parenchymal cells be proportional to flow.

1.3.3. *Parameter tools*

Parameter Evaluation — In certain instances, it may be useful for several parameters to have the same numeric value (e.g., the capillary permeability for the three tracers). Parameter expression evaluation can link the value of “slave” parameters to that of a “master” parameter. Changing the value of the master automatically changes the values of the slaves. Note: a parameter, including scalars, may not be slaved onto another parameter that is already slaved to a parameter or constant.

Two types of parameter tools are provided in MMID4: The eval field for each parameter, and a set of general scalars. The former allows a parameter to be set to equal a function of one or more other parameters.

Parameter scalars — Although MMID4 has a wide range of input parameters, situations arise in which a user wants to use a parameter that is not provided by the program. General scalars are provided for this purpose. These scalars allow the user to create a new result parameter that is a linear combination of other parameters. The set of scalars is simply a set of parameters that is not tied to the model, but that can be set, like any other parameter, either by entering a value directly or by slaving the scalar to another parameter in the Eval field.



e_PSg is slaved to p_PSg

2. MMID4 TUTORIAL

2.1. Getting Started

Conventions used in this tutorial

This tutorial assumes you are running MMID4 (and XSIM) under the UNIX operating system with a windowing environment, and using the C-shell as your command interpreter. If you are not using the C-shell consult your system manager or another knowledgeable person about procedures given below that do not work as you expect. The commands and text you are to enter are shown in a monospaced (`monospaced`) font. Unless otherwise specified (or evident from the context), commands are entered at the system prompt: no representation of the return or enter key is used.

XSIM notes

While this tutorial is not intended to teach you the details of XSIM, it is sometimes necessary to include information specific to XSIM. Such information follows a "XSIM Note" heading such as is shown in the example below:

XSIMNote: Text of XSIM-specific information. This usually includes a reference to the applicable section of the XSIM User Guide.

Exercises

Exercises follow headings of the kind exampled below:

Exercise: Title (Parameter file NAME.par)

"Title" is a short descriptive title of the exercise, and NAME is the name of the parameter file you should select from the tutorial database.

Appendix B contains samples of the graphical results that you should obtain by running the exercises.

Menu shorthand

The path to many windows or parameters is often abbreviated in this manual. For example, the statement "Change *Parameters: Input functions* > *Vascular input* ^*Function* to Sine pulse train" is a shorthand way of describing how to change the vascular input to a sine pulse train. A full description would read: "Click on the Parameters menu, then select the Input functions from the pop-up menu, and then select the Vascular inputs option. Now, in the Vascular Input window click on the Function button and select Sine pulse train." Fig. 2.1 provides a graphical example of this operation.

An explanation of the shorthand notation: The first word in the series is always one of the pull-down menus in the main window. The colon before the second phrase ("*: Input functions*" in this case) indicates that this is a pull-down menu option. The right angle bracket before a phrase ("> *Vascular input*"), designates an option on a secondary pop up menu. Finally the carat (^) preceding a word or phrase ("^*Function*") indicates the name of a button or field in the newly opened window. Note that menu names are printed in *emphasis* in this manual.

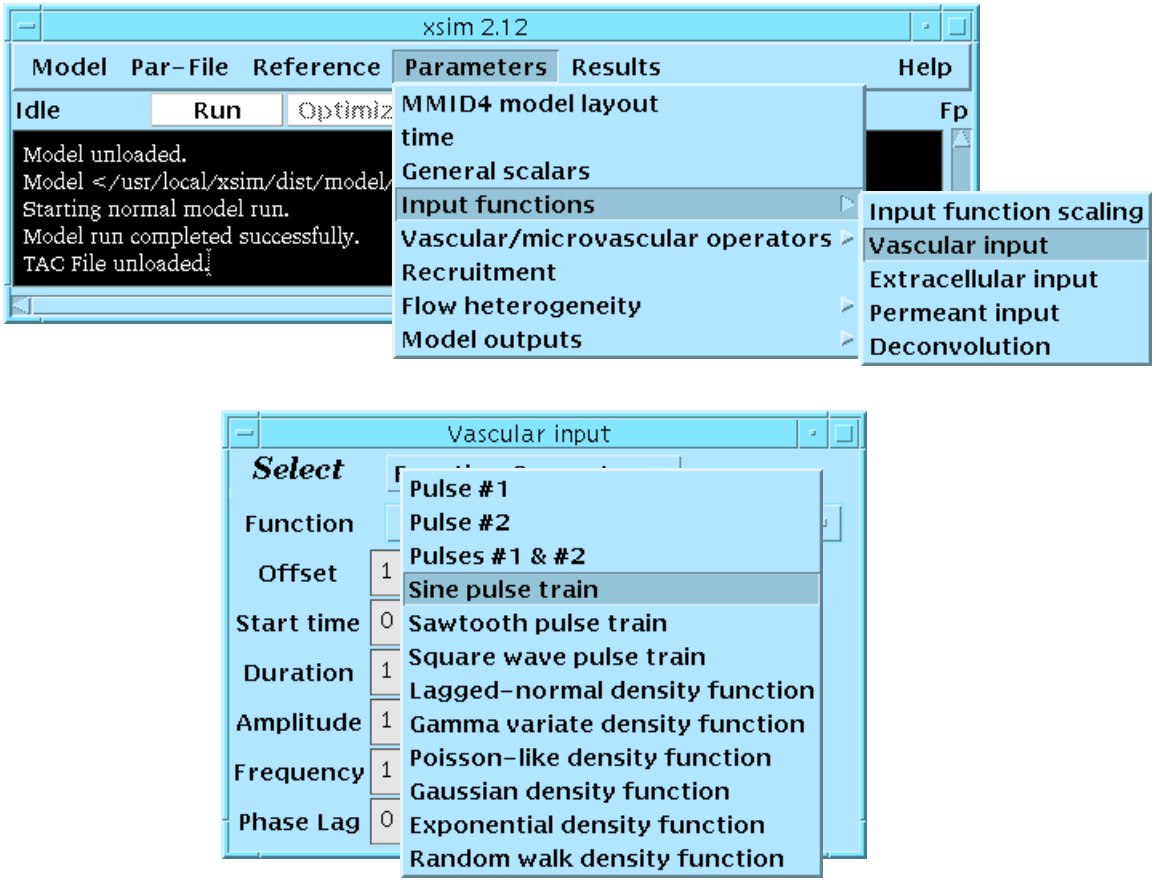


Figure 2.1. Opening the vascular input window from the XSIM main window (top) and selecting the function option sine pulse train from the opened window (bottom). The shorthand for this operation is: Change *Parameters: Input functions > Vascular input ^Function* to Sine pulse train.

2.1.1. *Overview*

In this section, “Getting Started”, you will learn how to set up an environment for the simulation interface that is used by MMID4. You will also learn how to start MMID4 and load values for its parameters from disk files.

2.1.2. *The XSIM simulation interface*

MMID4 runs under the XSIM simulation interface. Thus, it is necessary to become acquainted with XSIM before attempting to use MMID4.

XSIM is a graphical simulation control interface that runs in a UNIX windowing environment. It was developed by the National Simulation Resource (NSR) to run computer simulations of various types; its functions and usage are described in the XSIM User Guide. The appearance of the XSIM screen is not predefined, but is assigned at run time according to a user-defined configuration file. This file specifies the parameter values and their locations in the pop-up windows, as well as the content and appearance of the windows.

Upon starting MMID4, the MMID4 model layout window appears. The coordinates and colors of the text, buttons and graphics in this window are all specified in the configuration file.

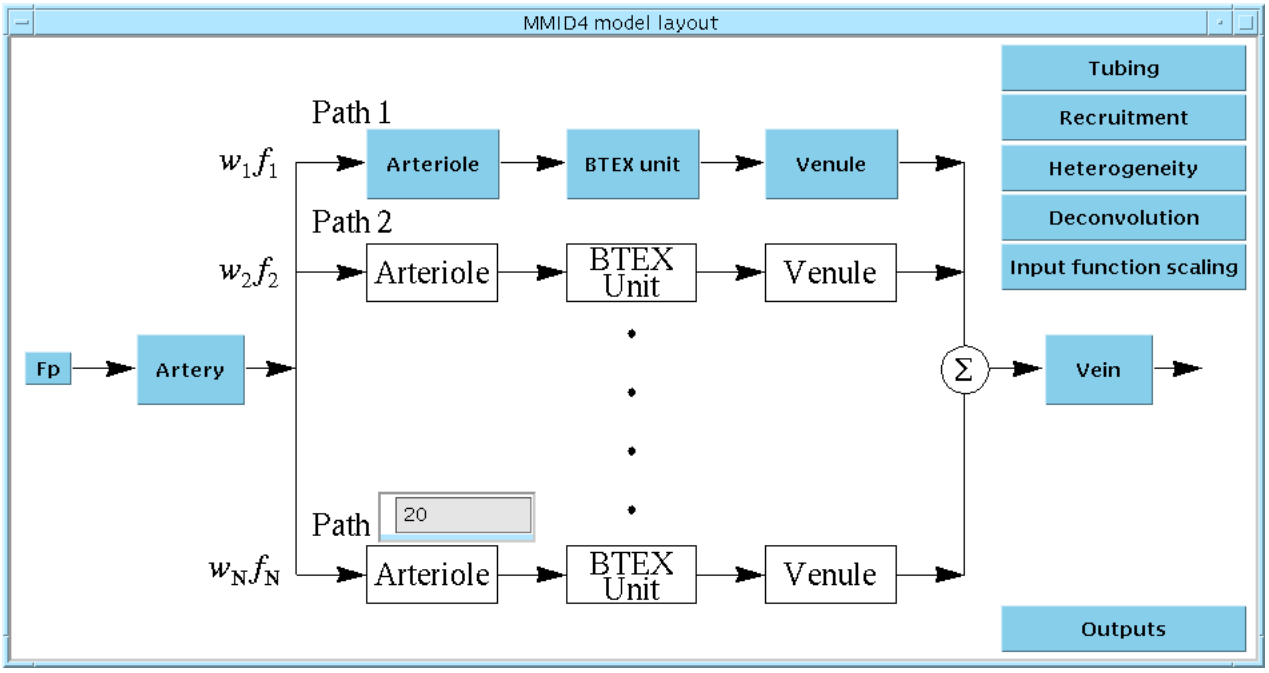


Figure 2.2. The MMID4 model layout window.

2.1.3. *Setting up a XSIM environment*

If XSIM has been fully installed on your system, the environment should be properly configured for running the MMID4 tutorial. If changes are required, section 2.1 of the XSIM Interface Reference Manual gives complete information about setting up a custom environment.

2.1.4. *Running the MMID4 model*

Running MMID4 with the tutorial parameter files

To run the exercises of this tutorial, you should use the MMID4 tutorial parameter files. To do so, you must first copy them into your directory. Use the following command sequence.

```
cd
mkdir MMID4tutor
cd MMID4tutor
zcat XSIMHOME/dist/model/MMID4tutor.tar.Z |tar xf -
```

Where *XSIMHOME* is the XSIM installation directory, usually `/usr/local/xsim` (check with your system administrator).

This tutorial database has a number of MMID4 parameter files that have been preloaded with the values appropriate to each exercise. Note that this also

copies the reference data files that are used in some of the tutorial exercises. You are now ready to run the model. Enter

```
xsim -m mmid4 -p 1input.par
```

to load the first tutorial parameter file. The name of each tutorial file is compounded from the tutorial number and a descriptive name, and terminated with the '.par' extension.

Note that each time you want to run one of the tutorial exercises you should use the command `cd ~/MMID4tutor`. This will place you in the subdirectory where the tutorial parameter and reference data files are located.

Running MMID4 for general usage

If you have previously run MMID4, you can select a parameter file that was previously used for MMID4; otherwise, you have two options to load meaningful values into the selected parameter file.

1. Import the values from a another user's parameter file.
2. Modify the default values for all the relevant parameters manually.

The second option is not usually recommended because many parameters need to be entered. It is nearly always better to load parameters from another user's parameter file and then modify the values required to make it appropriate for your needs.

In order to create your own parameter file, select *Save as...* from the *ParFile* pull-down menu in the main window. Enter a name (e.g. test.par) for the parameter file in the Parameter File field, then press return.

2.1.5. *Summary*

In going through this part of the MMID4 Tutorial, you set up an XSIM environment that allows you to run MMID4 and any other model that uses XSIM. You made a copy of the MMID4 tutorial directory that you can use and modify when you run the exercises in the remainder of the tutorial. Additionally, you learned how to start MMID4 and have it load parameter values from your parameter files or from the tutorial parameter files.

2.2. Basic Exercises

2.2.1. *Overview*

In this section you will learn some of the ways available to specify an input function for the model. You will learn then how to set up the basic parameters required to configure MMID4. Having these two items in hand, you will be able to run a simulation using MMID4 and will learn how to select results for display.

2.2.2. *Generating an input function*

Introduction

In this exercise, you will examine some of the functions that are available from the function generator in MMID4. A summary of the types available is listed in Table 2.1; the types range from simple pulses to a range of probability density functions (PDFs). Another method of generating an input function, deconvolution of the vascular tracer output curve, is discussed in Section 2.5.3.

Table 2.1: Available input functions

Pulses
Single pulse #1 or #2
Double pulses
Sine wave pulse train
Sawtooth pulse train
Square wave pulse train
Probability densities
Lagged normal
Gamma variate
Poisson-like
Gaussian
Exponential
Random walk

Three of the input functions provided deserve special consideration. The random walk, gamma variate, and lagged normal density functions have been shown to provide reasonable descriptions of concentration curves obtained from actual indicator dilution experiments in which the indicator remained in the vasculature. Sheppard (1954) fit random walk density functions to experimental data obtained from isolated, perfused canine heart, lung (without the heart), and liver experiments. Thompson et al. (1964) examined the utility of the gamma variate density function to fit peripheral arterial dye dilution cardiac

output curves in patients. In these experiments, injections were made into the right atrium or pulmonary artery, and dye concentrations were measured by continuously sampling peripheral arterial blood through a cuvette densitometer. The lagged normal density function was examined as a model for arterial dilution curves in man by Bassingthwaighte et al. (1966). These investigators sampled curves in a peripheral artery following injection of dye into either the superior vena cava or the aorta. All three of these distributions provide good fits to the concentration-time curves of indicator in the arterial system. They can, therefore, be used as an approximation of the form of the input function to the target organ following a bolus injection.

Exercise: Input functions (Parameter file 1input.par)

Start MMID4 with parameter file 1input.par of the tutorial database selected:

```
xsim mmid4 -p 1input.par
```

Select *Plot Area 1* from the *Results* pull down menu, then click on the Run button in the main window. The parameters have been set to select a step function that starts at one second and has a four second duration. One result, the value of the input (*Cin_v*), is plotted in the Plot Area 1 window.

XSIM Note: XSIM plots simulation results with open symbols. Thus when a square wave is plotted, you see only the high and low levels with no connection between them. You can direct XSIM to use different colors and symbol sizes by clicking in the desired parameter box and then clicking on a color button or one of the three symbol size buttons.

Summary Using the function generator is only one method of obtaining an input function for MMID4. This is the method that will be used for the Basic Exercises, Indicator Dilution Principles Exercises, and most of the Advanced Exercises. Details of generating input functions in MMID4 are given in Section 3.2. You should read that section carefully before using MMID4 for modeling and data analysis, because the number of options is large and some choices of parameter values interact with others.

2.2.3. *Configuring the model*

Introduction In this exercise you will set the parameters that control the basic configuration of the vascular units of MMID4. These include the mean blood flow, the number of flow paths, the volumes and relative dispersions (*RD*) of the nonexchanging vessels, and the transport and exchange parameters of the blood–tissue exchange (BTEX) units.

Exercise: MMID4 configuration (Parameter file 2config.par)

XSIM Note: You can load the values from parameter file 2config.par by leaving XSIM, restarting MMID4, and selecting parameter file 2config.par as the file to be used. If, however, you are already running MMID4, you can load the values from parameter file 2config.par without leaving the program. Pull down the *Par-File* menu to select *Load...*, and click on ‘No’ in the popup window that

appears. In the Load Parameter File window that appears, either type 2config.par in the parameter file field and then click on the load button, or double click on 2config.par in the scrolling Files menu.

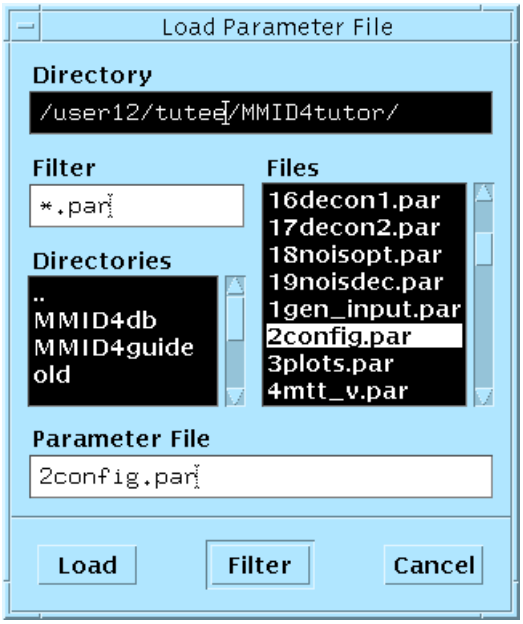


Figure 2.3. Load Parameter File window.

The mean plasma flow

The mean plasma flow, F_p, is set in the MMID4 model layout window. Move your cursor over the *Fp* button to see the current value of mean plasma flow, and note that the value disappears when the cursor moves away from the button. Now double click on the *Fp* button in order to open the parameter control window for *Fp*. Note that its units are ml g^{-1} min^{-1}.

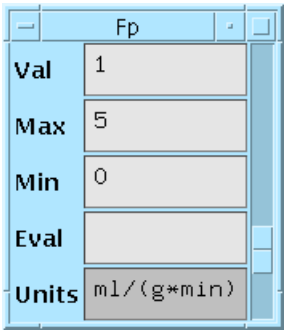


Figure 2.4. Parameter control window for flow, *Fp*

The number of flow paths

As shown in Fig. 2.5, MMID4 contains a set of parallel flow paths. The number of paths used is also set in the MMID4 model layout window. It must be in the range 1–20.

The nonexchanging vessels

The nonexchanging vessels are the input tubing, artery, arterioles, venules, vein, and output tubing shown in Fig. 2.5. Including these vessels allows you

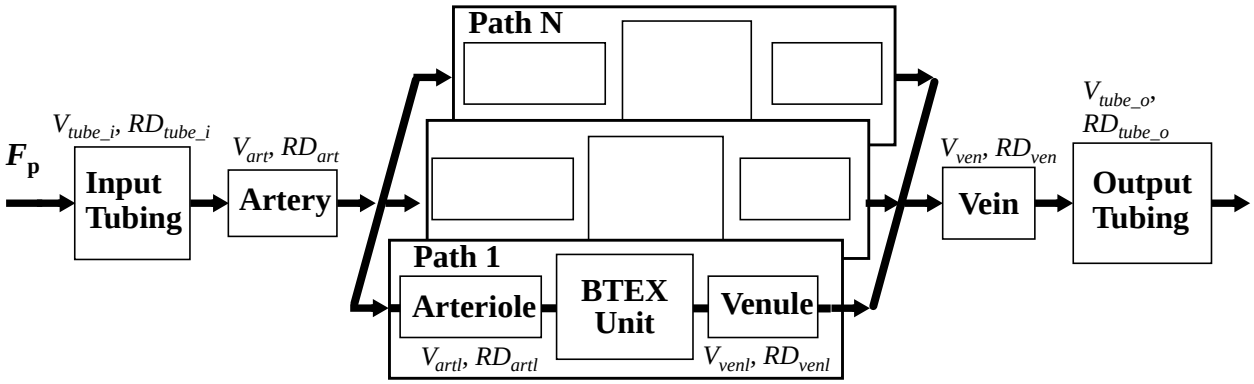


Figure 2.5. Arrangement of vascular units in MMID4. There is only one input tubing, artery, vein, and output tubing. The model contains N parallel flow paths each containing one arteriole, blood–tissue exchange (BTEX) unit, and venule.

to examine the influence of vessels that do not allow exchange with the tissue but do contribute to tracer delay and dispersion. When using these vessels, you must define their volumes (ml/g of organ) and the relative dispersion of the vessel. The windows containing lists of the parameters of all the nonexchanging vessels are also shown in Fig. 2.6. Note that the values set for arteriole and venule parameters are used for all flow paths.

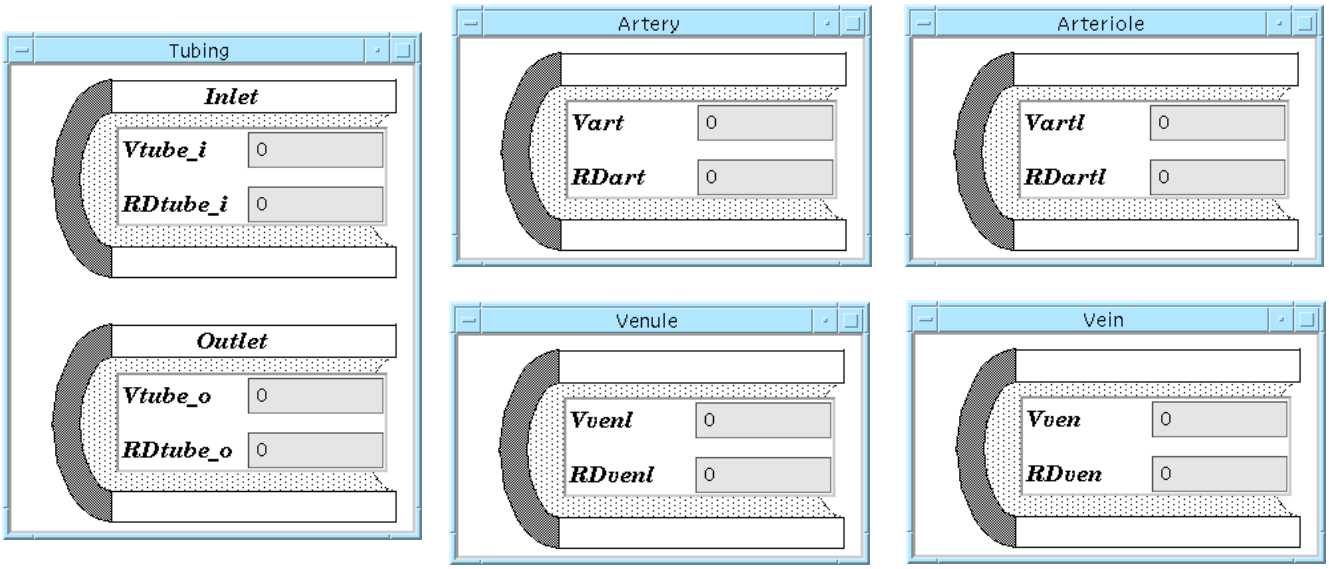


Figure 2.6. Nonexchanging vessel parameters.

If the volume of a nonexchanging vessel is set to zero, that vessel is not included in the modeling. The exercises of this tutorial do not use the nonexchanging vessels, thus all their volumes should be set to zero.

The blood–tissue exchange (BTEX) units

The effects of the BTEX units are controlled by their volumes (V), membrane conductances (permeability-surface area product, PS), axial diffusion coefficients (D), tracer consumptions (G), and initial concentrations (C_0). Since

MMID4 models three tracers (intravascular, extracellular, and permeant), there are three sets of these parameters. Fig. 2.8 shows the diagrammatic location of these parameters, and Table 2.2 shows the definitions, units, and typical values for the parameters. For this exercise the initial concentrations, consumptions and diffusion coefficients will be set to 0.0 for all tracers.

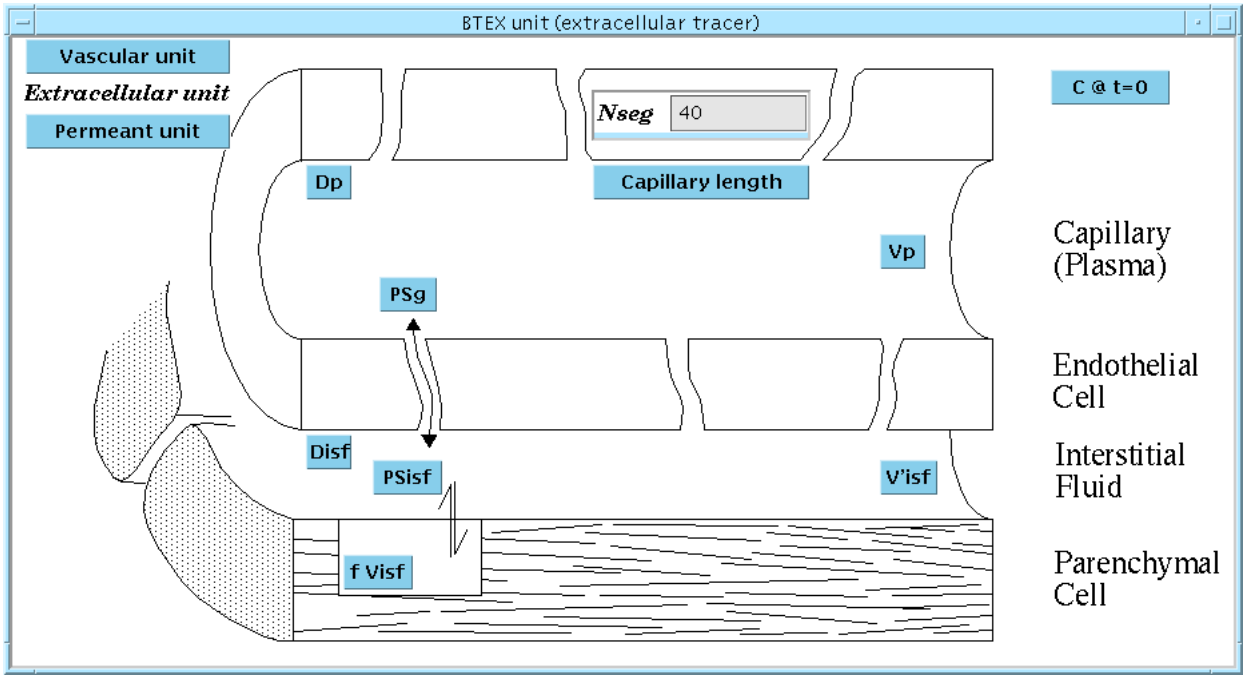


Figure 2.7. Extracellular tracer BTEX unit window.

To view the BTEX parameter values, click on the BTEX unit button in the MMID4 model layout window. The vascular unit window will open up. Click on the Extracellular unit button. The second interstitial fluid region is not used in this exercise; thus fV_{isf2} and PS_{isf} are both set to 0.0.

Display the values of the permeant tracer parameters by clicking on the Permeant unit button. The second ISF region is not used for this tracer either. In addition to setting fV_{isf2} and PS_{isf} to zero, PS_{pc2} is also set to 0.0.

In addition to the parameters that are specific to each tracer, four BTEX parameters are common to all the tracers. Plasma flow (F_p), plasma volume (V_p) and number of segments (N_{seg}) determine the internal time step used to evaluate the BTEX units ($dt_{internal}=V_p*60/(F_p*N_{seg})$). Because the time required for solving the model is proportional to the square of N_{seg}, choosing the number of segments is a compromise between solution time and accuracy. For these exercises, N_{seg} has been set to 10.

The final common parameter is the capillary length, l_{cap}, in centimeters. It is usually set to 0.1, but, since axial diffusion is not used in the tutorial exercises, its value will have no effect.

Running the model

Having set the mean plasma flow, the number of flow paths, and the parameters for the nonexchanging vessels and the BTEX units, you can now run the model

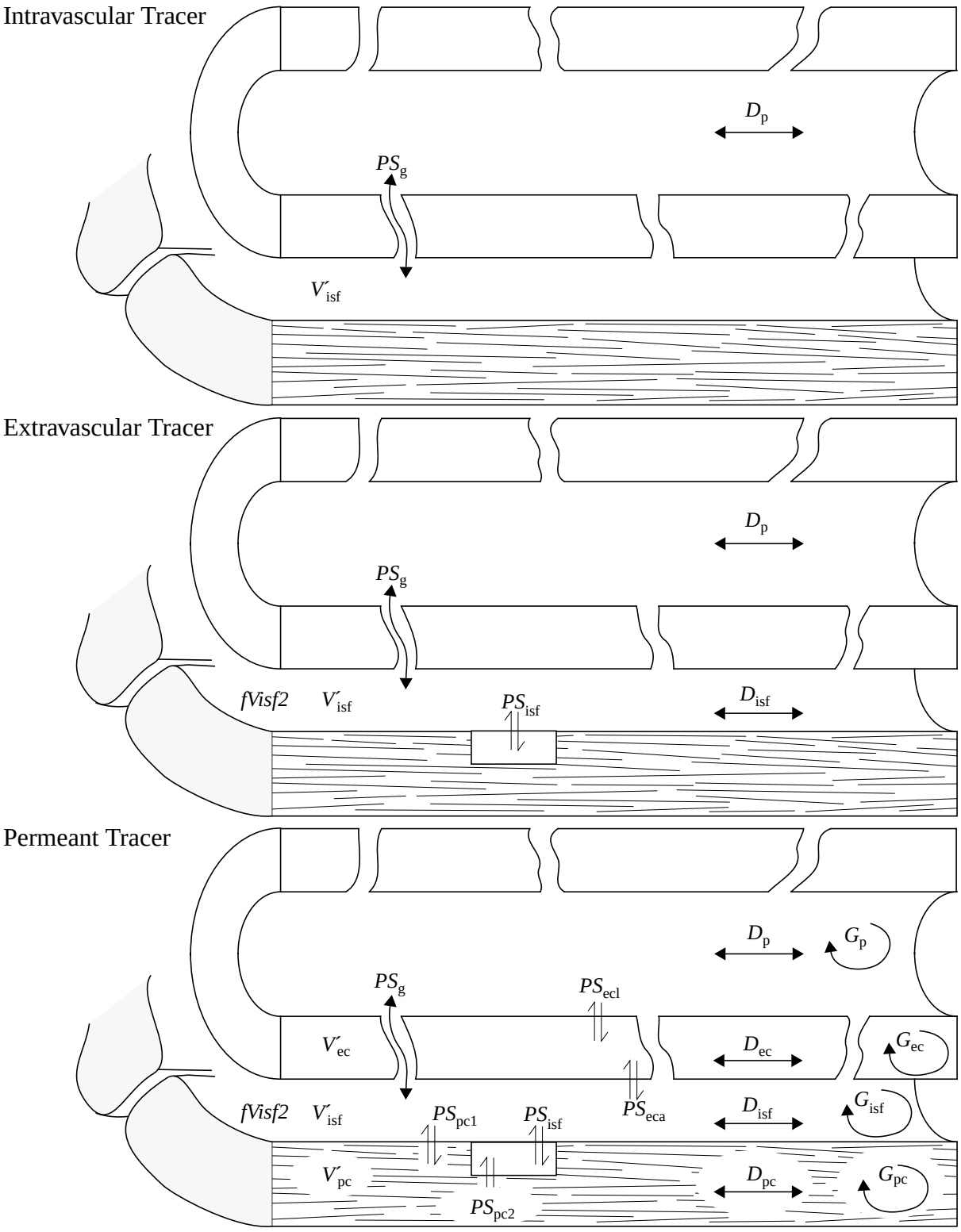


Figure 2.8. Upper: Parameters that define the intravascular tracer. *Middle:* Parameters that define the extracellular tracer. *Lower:* Parameters that define the permeant tracer. Symbols are defined in Table 2.2.

Table 2.2: MMID4 blood–tissue exchange parameters

Symbol	Definition	Units	Typical values			
			All	Intra-vascular	Extra-cellular	Permeant
F_p	Plasma flow	ml/(g · min)	1.0–2.0			
V_p	Plasma volume in exchange unit	ml/g	0.02–0.04			
N_{seg}	Number of axial segments	NONE	1-60			
l_{cap}	Capillary length	cm	0.1			
C_0	Initial concentration in all regions	mg/ml		0.0	0.0	0.0
PS_g	*PS* of capillary (endothelial gap)	ml/(g · min)		0.0	1.0	1.0
PS_{ecl}	*PS* of the luminal side of endothelial cell	ml/(g · min)				0.25
PS_{eca}	*PS* of the abluminal side of endothelial cell	ml/(g · min)				0.25
PS_{isf}	*PS* for ISF1 to ISF2 diffusion	ml/(g · min)			0.0	0.0
PS_{pc1}	*PS* of parenchymal cell to ISF1	ml/(g · min)				1.0
PS_{pc2}	*PS* of parenchymal cell to ISF2	ml/(g · min)				0.0
V'_{isf}	Total volume of distribution in ISF space	ml/g		0.0	0.15–0.25	0.15–0.25
fV_{isf}	Fraction of V'_{isf} in ISF2	NONE				0.0
V'_{ec}	Volume of distribution in endothelial cells	ml/g				0.01–0.02
V'_{pc}	Volume of distribution in parenchymal cells	ml/g				0.6
D_p	Axial diffusion in capillary (plasma)	cm^2/s		10^{-4}	10^{-4}	10^{-4}
D_{isf}	Axial diffusion in interstitial space	cm^2/s			10^{-5}	10^{-5}
D_{ec}	Axial diffusion in endothelial cell	cm^2/s				10^{-5}
D_{pc}	Axial diffusion in parenchymal cell	cm^2/s				10^{-5}
G_p	Consumption in plasma	ml/(g · min)				0.0
G_{isf}	Consumption in interstitial space	ml/(g · min)				0.0
G_{ec}	Consumption in endothelial cell	ml/(g · min)				0.0
G_{pc}	Consumption in parenchymal cell	ml/(g · min)				0.0

by clicking on the run button. The input function that has been selected in this parameter file is a lagged normal density curve (LNDC). You will see it plotted along with outflow concentrations of the intravascular, extracellular, and permeant tracers. (Note that the same input curve is used for each of the three tracers.) Since all of these curves are plotted with small symbols, it helps to know something about the vascular system being modeled to know which is which. In this case, the first curve is, logically, the input curve and the remaining curves are the outflow curves. Which is the vascular tracer outflow curve? Since this tracer is confined to the vascular volume and the others diffuse out into the tissue, you can conclude that it is the outflow curve with the shortest transit time and the highest peak. Since the permeant tracer diffuses into ISF and cells, its curve has the lowest peak and longest transit time; the remaining curve is the extracellular curve.

Summary

In this exercise you have loaded a set of MMID4 parameters, checked the values that govern the behavior of the vascular units, and run the model. A set of results curves were plotted. Knowing what results were plotted, you used some knowledge about the nature of the tracers and the vascular system to associate a specific output curve with the appropriate tracer. The term *transit time* was introduced; this will be discussed further in an upcoming exercise.

2.2.4. *Selecting results for display*

Introduction

MMID4 has several types of results that can be plotted. Of primary interest are the inputs, the outflows, and the tracer contents. In this exercise, you will focus on plotting the outflows and tracer content in the vascular system. To begin, load parameters from parameter file 3plots.par by switching to a new parameter file (*Par-File: Load...*) or by starting MMID4 with parameter file 3plots.par.

Exercise: Plotting results (Parameter file 3plots.par)

This parameter file has been preset to use a LNDC input for all tracers. Run the model and note that two curves are plotted: the input and the outflow of the intravascular tracer.

XSIM Note:

There are two methods to specify the plotted curves. Either directly type in a parameter in a ‘Y Parameters’ field, or open the pick control window by right clicking in the ‘Y Parameters’ field, then scroll to and click on the desired parameter in the list. To change the color or style of the curve, click in the ‘Y Parameters’ field of interest, then click on either a color or size button just above the graphing area at the right hand side.

For this exercise, the two curves plotted are the vascular input and vascular outflow. (The extracellular and permeant input curves are identical to the vascular input.) The size and axis scaling of the plot are under user control. In this data file, values have already been specified.

Table 2.3 shows the parameter numbers associated with the input, outflow, and tracer contents curves of the three tracers. Display the permeant outflow by

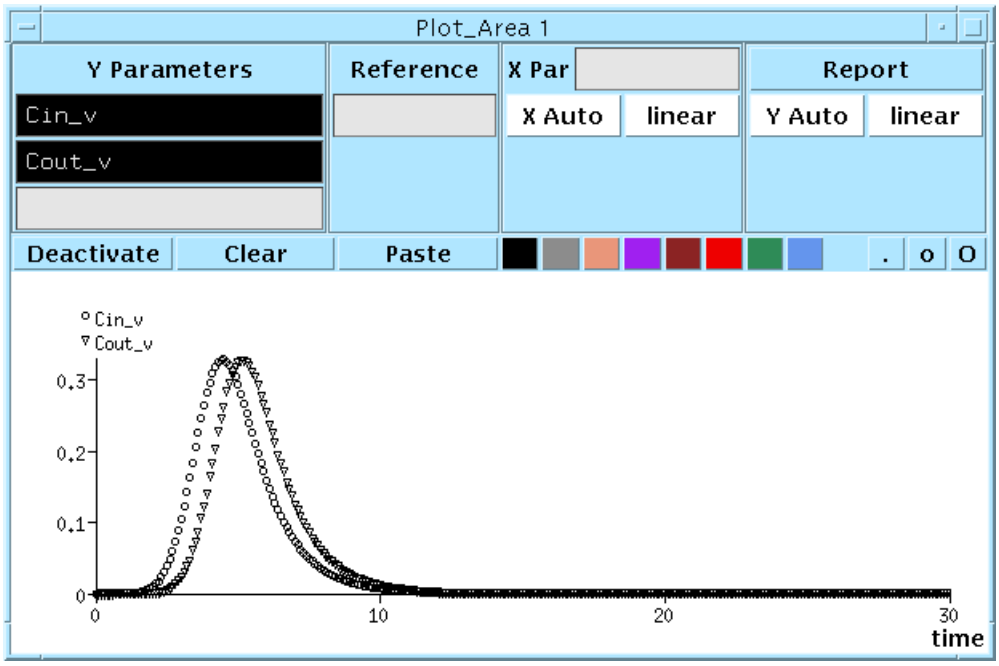


Figure 2.9. The Plot Area 1 window with input and output concentration curves.

typing `Cout_p` in the third parameter field. Run the model again, and note the additional curve.

Table 2.3: Parameters for tracer input, outflow and content curves

Symbol	Parameter usage
Cin_v	Intravascular input
Cin_e	Extracellular input
Cin_p	Permeant input
Cout_v	Intravascular outflow
Cout_e	Extracellular outflow
Cout_p	Permeant outflow
Q_v	Intravascular content
Q_e	Extracellular content
Q_p	Permeant content

Now, add the tracer content for the vascular and permeant tracers (*Q_v* and *Q_p*) to plot area 1 and run the model again. You should have five curves displayed. The tracer contents curves are near the axis as their scale is quite different than that of the outflow curves.

XSIM Note: There are two ways to change the scale used for plotting simulation results. The y-axis scale can be set manually or autoscaling on the largest plotted parameter can be specified. If the maximum values of the curves differ greatly it is usually better to put curves that require a separate scale in another plot area.

Move the content curves to the second plot area by selecting *Results: Plot Area 2*; then entering *Q_p* and *Q_v* in the parameter fields. Remove the content curves from the first plot area by clicking on the field containing *Q_v*, then clicking on the 'Deactivate' button. Do the same for the *Q_p* field. Run the model again. Does this scaling make the curves easier to see? Which is the curve for the intravascular tracer? (Remember that the intravascular tracer has access to a smaller volume than the permeant tracer, and, thus, leaves the BTEX unit more rapidly.)

Summary In this exercise you learned about the XSIM plot parameters, and about adding or deleting graphical output curves. You were also introduced to XSIM plot areas and scaling of simulation output.

2.2.5. *Summary*

In these basic exercises, you have learned how to specify an input curve that is generated by the function generator. You have also examined, and perhaps altered the MMID4 parameters. Finally you were introduced to some of the results parameters that are available in MMID4 and learned how to specify which results are plotted. You are now ready to move on to exercises that deal with basic indicator dilution principles using MMID4.

2.3. Basic Indicator Dilution Principles

2.3.1. *Overview*

The exercises in this section introduce the concepts of *mean transit time* (i.e., the average time taken by a tracer particle to pass through the system) and *membrane conductance* (i.e., the membrane permeability-surface area product, *PS*). You will use the model to calculate mean transit time and membrane conductance. With these basic principles in hand, you will examine the effects of the presence of cells and of the input function shape on the outflow curves. Finally, you will examine some factors that complicate the assumptions that underlie these basic principles and complicate the interpretation of indicator dilution curves.

For these exercises, you will run the model in its simplest state (i.e., using one pathway without the nonexchanging vessels).

2.3.2. *Mean transit time*

Introduction

Indicator dilution techniques are often used to determine the mean transit time of a solute through a vascular bed. The mean transit time, $\bar{t}$, of a tracer depends on the volume of distribution of the tracer and the flow through the exchange unit:

$$\bar{t} = V / F \text{ seconds}, \tag{2.1}$$

where V is the total volume of distribution of the tracer in the vascular bed and F is the total flow. In the exercises of this section, you will examine the mean transit time of the intravascular and extracellular tracers.

Exercise: Mean transit time of the intravascular tracer (Parameter file 4mtt_v.par)

Parameter file 4mtt_v.par has been set up to examine the mean transit time of the intravascular tracer. Load parameter file 4mtt_v.par. The input function selected is a lagged normal density curve (LNDC) with an area of 1.0, and a mean of 2.0 seconds; the intravascular tracer has been selected as the first output to be plotted. Examine the values of the parameters that govern the intravascular tracer mean transit time (i.e., total plasma flow, *Fp* at *Parameters: MMID4 model layout ^ Fp*, and plasma volume in the BTEX window, Fig. 2.10, found at *Parameters: vascular/microvascular operators > tissue-capillary units > BTEX unit(vascular tracer) ^ Vp*) and calculate the expected mean transit time. (NOTE: Be careful of units. Flow is given in ml g^{-1} min^{-1}, and you want *t* in seconds.) Run the model. The mean time of the first output plotted is calculated by XSIM and stored in the report at *Results: Plot Area 1 ^Report*.

Examine the xmean value of the *Cout_v* parameter. Does it have the value that you predicted? The calculated mean time of the outflow curve is the $\bar{t}$ of the input function plus the *t* of the tracer in the exchange unit. Remember that the mean of the LNDC input is set to two seconds (*Parameters: input func-*

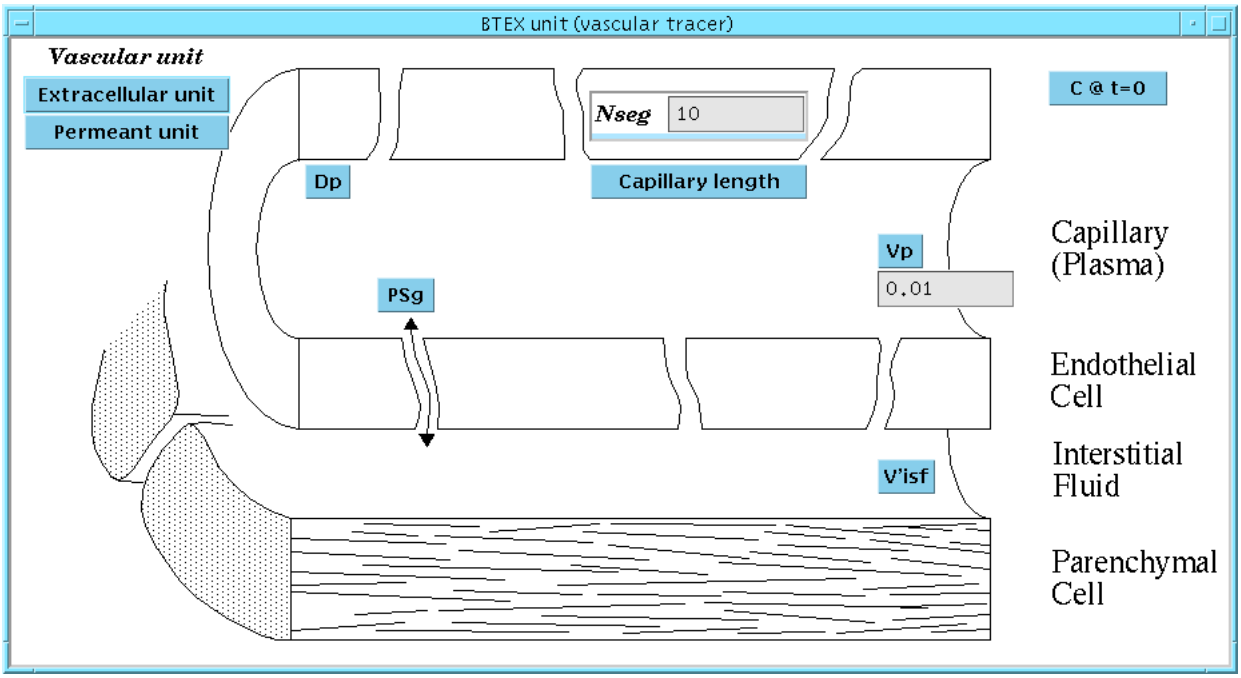


Figure 2.10. Vascular tracer BTEX unit window.

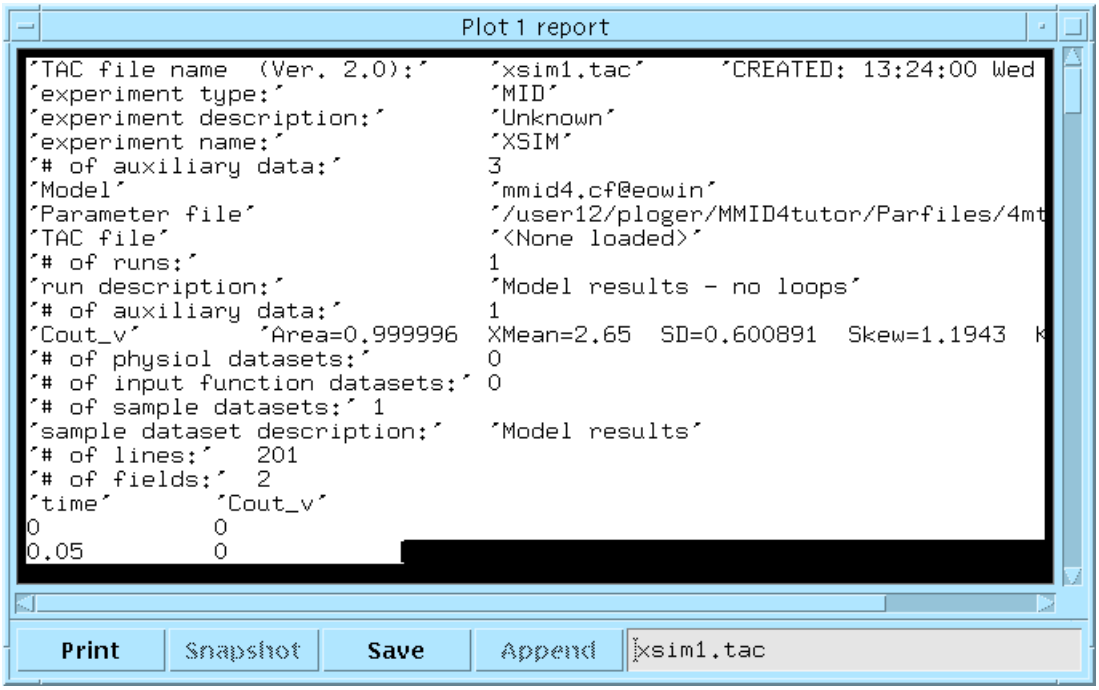


Figure 2.11. Plot Area 1 report window

tions> vascular input ^mean). When the mean time of the output curve is corrected for the input transit time, how well does the result agree with your prediction? A common problem with the BTEX operators is that their output is delayed by one time step; this adds one Δt to the calculated transit time. Cor-

rect XSIM's calculated value for the simulation time step (*Parameters: time ^ Incr*). How well does this agree with your prediction now?

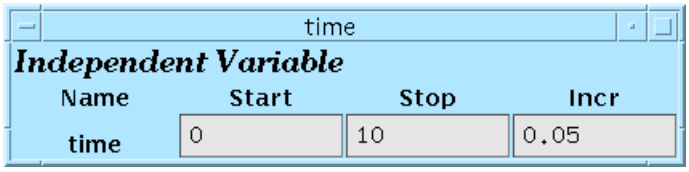


Figure 2.12. Time window

You should now be able to predict the xmean value of *Cout_v* for a given flow and volume. Choose new F_p and V_p values; run the solution and check your prediction against XSIM's calculation.

Summary

The mean transit time, $\bar{t}$, is dependent on the volume and flow of the vascular bed. The mean of the outflow curve must be corrected for the mean of the input curve and the simulation time step to give the correct answer. (N.B. The time step correction is a property of the BTEX operators and not a property of the vascular system.)

Exercise: Mean transit time of the extracellular tracer (Parameter file 5mtt_e.par)

This exercise is similar to the previous one except that you will examine the results for the extracellular tracer.

Start this exercise by loading parameter file 5mtt_e.par. Examine the parameters that govern the extracellular tracer, which are located on both the MMID4 model layout window and at *Parameters: vascular/microvascular operators > tissue-capillary units > BTEX units(extracellular tracer)*. Note that capillary wall conductance, PS_g, has been set to 1.0 ml g^{-1} min^{-1}; thus, the tracer will enter both plasma and interstitial fluid (ISF) spaces. Calculate the $\bar{t}$ of the tracer in the exchange unit. (What is *V* in this case?) Run the solution. How does the value of $\bar{t}$ for this tracer (*XMean* of *Cout_e* corrected for input function and time step) compare to what you calculated? Extend the solution time by changing *Parameters: time ^stop* to 50 seconds; run the model again. Has the $\bar{t}$ derived from the outflow concentration curve changed? Why? The area of the input function is 1. How much of the outflow curve have you seen? (Look at the calculated area of the curve in *Results: Plot area 1 ^ report*.) How well have you recovered what you put in? Increase the solution time to 100 seconds. How does the result compare now? Why?

Summary

The mean transit time of a vascular bed, as measured by indicator dilution, is determined by the flow and the total volume accessible to the tracer. It is important to remember that accurate estimates of the mean transit time of tracers require good recovery of the outflow concentration curve. Thus, be sure to run the model for long enough to let almost all of the tracer exit from the system.

2.3.3. *Membrane conductances*

Introduction

Indicator dilution techniques may be used to obtain estimates of the conductance of the endothelial membrane for various solutes expressed as the permeability-surface area product (*PS*). Crone (1963) described a technique for estimating the *PS* by determining the extraction of the tracer during its initial pass through the tissue. Assuming no back-diffusion, Crone derived the following expression:

$$PS = -F \ln(1 - E), \tag{2.2}$$

where *PS* is the permeability-surface area product, *F* is the plasma flow, and *E* is the initial extraction of the tracer.

Exercise: Crone calculation of *PS* (Parameter file 6ps_crone.par)

In this exercise you will use the model to examine how well the Crone equation holds. Parameter file 6ps_crone.par is set up to use a lagged normal density function as the input, and plot the outflow concentrations of the intravascular tracer (*Cout_v*), the extracellular tracer (*Cout_e*), and the extraction of the extracellular tracer (*1-Cout_e/Cout_v*).

The extraction is defined as the fraction of tracer (relative to the intravascular tracer) that leaves the vascular space at any instant. It is calculated from the normalized concentrations of the reference tracer and a diffusible tracer:

$$E(t) = 1 - h_D(t) / h_R(t) \ , \tag{2.3}$$

where $h_D(t)$ is the normalized concentration of a diffusible tracer and $h_R(t)$ is the normalized concentration of the reference tracer. For this exercise, the reference is the intravascular tracer, and the diffusible is the extracellular tracer.

XSIM Note:

Note that expressions such as *1-Cout_e/Cout_v* can be entered into the y parameter field of the plot area windows. Expressions may consist of parameter names, arithmetic operators (+ - * / **), constants and mathematical functions. See Section 4 of the Interface Reference Manual for information on using expressions and appendix B, The XSIM Expression Evaluator, of the XSIM guide for a list of available constants and mathematical functions.

Run the model. You will notice that a series of error messages will print in the main window message area. Since the value of Cout_v is zero for the first 0.55 seconds, divide by zero errors occur. XSIM ignores these errors and sets the value of the equation to a very small number.

After the entire run is displayed, go to Plot Area 1 and click on the Report button. Obtain a mean value for the extraction of the extracellular tracer for several values occurring at the beginning of the extraction curve and use Eq. 2.3 to calculate the *PS* product. Check this value against the value of the PS_g for the extracellular tracer (*Parameters: vascular/microvascular operators > tissue-capillary units > BTEX unit(extracellular tracer) ^PSg*). How well do

they match? Over what range of *PS*'s will this work in the model? What about under real experimental conditions?

XSIM Note: All values plotted on the screen are also stored in TAC file format and are available for review by clicking on the Results button in the Plot Area window. These results are overwritten each time you run the model.

Repeat this exercise using the permeant tracer by replacing the outflow concentration of the extracellular tracer (*Cout_e*) with the permeant output concentration (*Cout_p*) in the extraction equation in the Plot Area 1 window. Vary the *PS* of the parenchymal cells. Can you detect these changes in your calculation? Vary the *PS* of the endothelial cell. Can you detect these changes in your calculation? What *PS* or combination of *PS*'s are being measured?

This technique assumes that the volume of distribution (V'_{isf} in the case of the extracellular tracer) is relatively large. For a particular PS_g (e.g., 1.0), progressively decrease the apparent volume of distribution for the tracer to examine what "relatively large" means in this context. Does this "definition" change if a different PS_g is considered (e.g., $PS_g = 5$)?

Summary The conductance of a membrane can be calculated from the extraction of a diffusible tracer using the Crone equation. The accuracy of the result is influenced by the relative values of the conductance and the volume into which the tracer distributes. When the tracer permeates more than one membrane, the results may be confounded by the physical arrangement of the membranes (e.g., membranes in series versus in parallel).

Exercise: Influence of *PS* on exchange (Parameter file 7ps_exch.par)

Introduction In this exercise you will examine the influence of membrane conductance on tracer exchange by performing three experiments. In the first, you will investigate the effect of capillary permeability (PS_g) on the amount of extracellular tracer that resides in the tissue. The second looks at the effect of the *PS*/*F* ratio on exchange, and the third examines the effect of *PS* on mean transit time. In doing this exercise, keep the following questions in mind:

1. Where does the tracer go?
2. When does the tracer appear in the outflow?
3. How does the extracellular tracer outflow curve compare with the reference (intravascular tracer outflow) curve?

Begin by loading the parameters from parameter file 7ps_exch.par. In this set of parameters, the number of axial segments (*Parameters: vascular/ microvascular operators> tissue-capillary units> BTEX unit(vascular tracer) ^Nseg*) is set to 10. After completing the exercise, you may want to go back and compare your results to those obtained when a smaller number of segments are used for the solution.

EXPERIMENT PS-1 For the first experiments, the input function parameters have been set to approximate a delta function input. To view the input function, pull down *Parameters* to *input functions*, then across to *vascular input*.

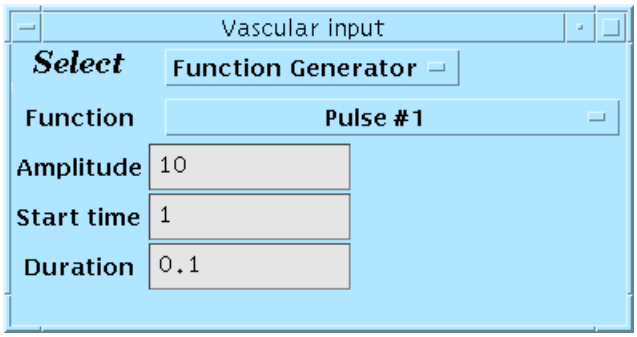


Figure 2.13. Vascular input window set for a pulse input function.

The plotting parameters have been set to plot the extracellular tracer quantity (*Q_e*) in plot area 1 and the intravascular tracer quantity (*Q_v*) in plot area 2:

PS_g for the intravascular tracer (*Parameters: vascular/microvascular operators> tissue-capillary units> BTEX unit(vascular tracer) ^PSg*) is set to 0.0 and for the extracellular tracer (*Parameters: vascular/microvascular operators> tissue-capillary units> BTEX unit(extracellular tracer) ^PSg*) to 0.01. Run the model. Are the shapes of the curves as you would have predicted? Remember, you are plotting the amount (mg/g) of tracer in the exchange unit (both intravascular and extracellular). Do the timing and shapes of the intravascular and extracellular curves differ? What does this tell you about the permeability of the capillary wall with respect to the extracellular tracer?

Next you will run the model to obtain a family of curves with progressively increasing PS_g for the extracellular tracer. Click on the *Model* menu and select *Run config.* In the Run Config window that appears, click on the Activate button to the right of the *Inner Loop* title. Make the parameter changes shown in Fig. 2.14 to put XSIM into the iterative mode and generate a series of six curves with PS_g ranging from 0.01 to 31.25.

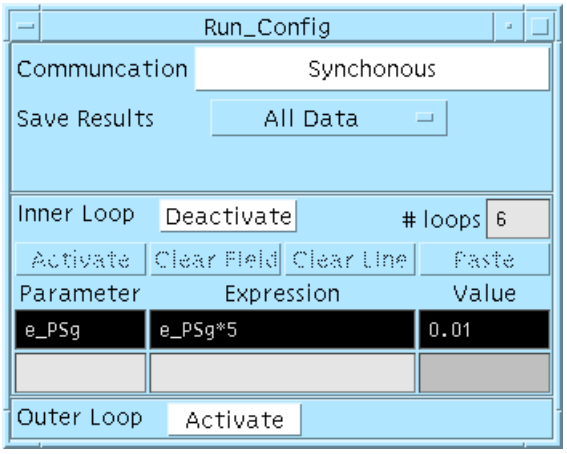


Figure 2.14. Run Config window with a single loop activated to run six times.

Run the model. As you increase PS_g, what happens to the amount of tracer in the interstitial space? Is this what you would have predicted? What would you predict the time course and shape of the outflow curves to be?

XSIM Note: XSIM's iterative execution mode allows the user to generate a set of solutions while changing the value of a number of parameters. The XSIM main window displays the value of the parameters as they change for each loop iteration.

Change the plot parameters to show the outflow curve for the extracellular tracer (*Cout_e*) in plot area 1 instead of the organ tracer contents. Click on the button labeled 'Y Auto' in the Plot Area 1 window to switch to manual y axis scaling. Enter 0.2 as a y axis maximum. Run the model again and observe the changes in the outflow and content curves. How do you explain the shape of the outflow curves? (It may be helpful to use a longer simulation time, e.g., 20 seconds, to get a more complete outflow curves.)

Summary With a low PS_g, the time course of the extracellular tracer in the tissue closely resembles that for the intravascular tracer. As PS_g is increased, more and more of the tracer diffuses into the ISF as the tracer bolus passes down the capillary. After the bolus exits the capillary, the tracer diffuses back into the capillary and is carried downstream by convection. This tracer may be carried out of the tissue or may diffuse back to ISF again. Looking at the tissue content curves, an early plateau is noted while the bolus is still in the capillary. This is followed by a monotonic decrease as the diffused tracer is carried out of the tissue. The outflow curves show an early spike of unextracted tracer followed by a tail that may decrease monotonically or may exhibit a peak depending on the value of PS_g.

EXPERIMENT PS-2 Return to the parameters used at the beginning of experiment PS-1 by reloading parameter file 7ps_exch.par

Change PS_g for the extracellular tracer (*Parameters: vascular/microvascular operators > tissue-capillary units > BTEX unit(extracellular tracer) ^ PSg)* to 30. Run the model for flows of 1 ml g^{-1} min^{-1} and 2 ml g^{-1} min^{-1}. (Set up a loop so you can see both curves on the screen.) Has increasing the flow significantly changed the amount of tracer reaching the interstitium? (HINT: The tail of the curve, especially just after the bolus exits the capillary, reflects the amount of tracer in the ISF.) By doubling the flow while maintaining a constant input function, we have introduced twice the quantity of tracer into the exchanging organ. How well has the amount of tracer entering the interstitium reflected this increase? This is an example of flow limited exchange.

Repeat the experiment with a PS_g of 0.5. In this case, how has increasing the flow changed the amount of tracer reaching the interstitium? Since the amount of tracer reaching the ISF seems to be independent of flow, what primarily limits the diffusion?

Summary The ratio of *PS* to *F* gives an indication of whether the limitation to exchange is primarily influenced by the barrier (*PS*/*F* small, barrier- or diffusion-limited) or flow (*PS*/*F* large, flow limited). When *PS*/*F* is 1, these factors contribute equally.

EXPERIMENT PS-3

Return again to the initial parameters of parameter file 7ps_exch.par. Change the input function to a lagged normal density function with the parameters shown in Fig. 2.15.

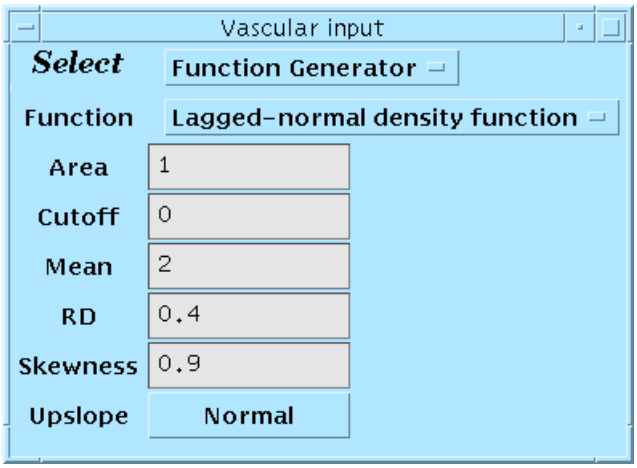


Figure 2.15. Vascular input window set for a LNDC input.

Set the display to show the outflow concentrations of the intravascular tracer (*Cout_v*) and the extracellular tracer (*Cout_e*) in plot area 1. Rescale the time axis by setting the stopping time to 30.0 (*Parameters: time ^Stop*). You may find it helpful to increase the vertical size of the plot area window by doing a click and drag on the upper or lower frame of the window.

Run the model to obtain a family of curves with increasing PS_g for the extracellular tracer as you did in Experiment PS-1 (e.g., PS_g in the range of 0.01 to 31.25). You should be able to predict the general shape of the extracellular tracer curves in relation to the intravascular tracer outflow curves. How well does your prediction match the model output?

Now turn off the loops by clicking the ‘deactivate’ button in the Run Config window. Set the value of PS_g to 10.0; run the model and examine the value of mean transit time in the Results window. Is it what you predicted? (Don’t forget to make the corrections for the input function mean and the time step.) Change the value of PS_g to 1.0 and check the calculated transit time again. Make sure that you allow the solution to continue long enough to see the whole outflow curve. (Keep increasing the stopping time, until the area of the curve, is at least 0.999.) Are the mean transit times for the two values of PS_g different? What determines mean transit time of a tracer? Change the value of PS_g to 0.1 and check the area and transit time calculations again. How large do you have to make the stopping time to recover 99.9% of the area? Why?

Now examine how the apparent volume of distribution for the extracellular tracer (V'_{isf}) influences these curves. For a given value of PS_g, decrease V'_{isf} (found in the BTEX unit (extracellular tracer) window) by half and repeat the experiment. Did the curves behave as you would have predicted?

Summary

The mean transit time is determined by the flow and volume only. While the presence of one or more permeable membranes will alter the shape of the out-

flow curve, the mean transit time is not affected. When *PS* is low, only a small amount of tracer escapes to the ISF; however, it also takes a long time for this tracer to re-enter the capillary and be carried out of the tissue.

2.3.4. *The effect of cells on indicator dilution curves*

Introduction

In this section, you will apply the principles reviewed above to predict exchange when cells are present. In these exercises you will continue to use MMID4 with just one pathway and use a lagged normal density function as the input function.

Exercise: Influence of the parenchymal cell (Parameter file 8p_vs_e.par)

In this exercise, you will focus on the permeant tracer and use the extracellular tracer as the reference. Load the values for parameter file 8p_vs_e.par. It has been initialized with the following parameters:

Parameter	Value
Extracellular Tracer	
V'_{isf}	0.15
PS_g	1.0
Permeant Tracer	
V'_{ec}	0.01
V'_{isf}	0.15
V'_{pc}	0.55
PS_{ecl}	0.0
PS_{eca}	0.0
PS_g	1.0
PS_{pc1}	1.0

Note that PS_g for the extracellular tracer is equal to that for the permeant tracer, all consumption parameters have been turned off (i.e., have a value of 0.0), and that the second ISF region has been turned off for both tracers. These parameters can either be accessed from the Parameters pull down and across menus, or by selecting *Parameters: MMID4 model layout* and clicking on the *BTEX unit* button. Choose the desired tracer from the buttons in the upper left hand of the BTEX unit window.

EXPERIMENT PC-1

Run the model with these initial parameters. The plot parameters are set to show the outflow concentrations for the extracellular and permeant tracers over a 60 second time period. How do these curves compare? Which is the curve for the permeant tracer? (Remember that it has a larger volume of distribution.) Are the differences primarily on the upslope, at the peak or at the tail of the

curve? Is this what you would predict? (It may be helpful to display the curves on a semi-log plot. You can do this by clicking on the button labeled ‘linear’ that is to the right of the ‘Y Auto/Y Manual’ button.)

Repeat the experiment using an iterative solution loop to run four solutions with PS_{pc1} ranging from 0.5 to 4.0. How does the permeant tracer curve change? Change the starting value of PS_{pc} to 4.0 and run the solutions again. Will further increases in PS_{pc1} change the curves? Why? (HINT: If PS_{pc1} becomes infinite, then the ISF and the parenchymal cell are effectively a single region with a volume of $V'_{isf} + V'_{pc}$.)

Reload the starting values and repeat the experiment once more, but this time plot the amount of extracellular tracer in the exchange unit (*Q_e*) and the amount of permeant tracer in the exchange unit (*Q_p*). Under these conditions, what does the difference between the two curves represent? Which curve is the permeant tracer? What effect does changing PS_{pc1} have on the residue function (i.e., the amount of tracer in the exchange unit)?

EXPERIMENT PC-2 Keeping the *PS* of the parenchymal cell constant (e.g., $PS_{pc1} = 1$), examine the effects of changing the apparent volume of distribution for the permeant tracer in the parenchymal cell by changing the value of V'_{pc}. Look at both the outflow curves and the residue functions as you did in Experiment PS-1. Which has a bigger effect on the curves, *PS* or *V*? Is this what you would have predicted?

Summary When the permeant tracer can diffuse across the parenchymal cell membrane, it has a larger volume of distribution than the extracellular tracer. This gives a permeant outflow curve with a lower peak and a longer tail that crosses the extracellular curve. The magnitude of the peak and time of the crossover are dependent on volumes and membrane conductances. The residual curves have complementary shapes.

Increasing the value of PS_{pc1} has a nonlinear effect since the parenchymal cell and ISF in effect become one region at high PS_{pc1}.

Exercise: Influence of the endothelial cell (Parameter file 9endoth.par)

In this exercise you will look at the combined effects of the endothelial cells and the ISF on the outflow curve and on mean transit time when tracer enters these regions in parallel or in series. The configuration of the model used for this exercise is shown in Fig. 2.16.

EXPERIMENT EC-1 The influence of the parenchymal cell has been removed, and the overall *PS* products and apparent volumes of distribution for the extracellular and per-

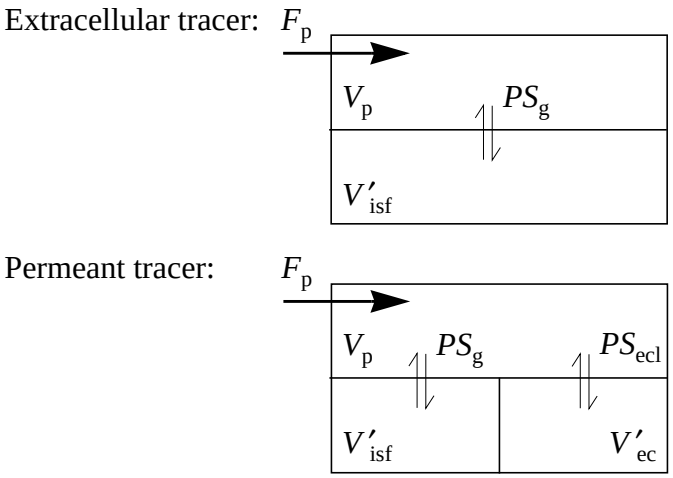


Figure 2.16. Configuration of the blood-tissue exchange regions for the extracellular (top) and permeant (bottom) tracers in Experiments EC-1 and EC-2. Note that the permeant tracer enters the ISF and endothelial cells in parallel.

meant tracers are equal. The model has been defined with the following characteristics:

Parameter	Value
Extracellular Tracer	
V'_{isf}	0.15
PS_g	1.0
Permeant Tracer	
V'_{ec}	0.1125
V'_{isf}	0.0375
V'_{pc}	0.0
PS_{ecl}	0.25
PS_{eca}	0.0
PS_g	0.75
PS_{pc1}	0.0

Note that the *total* membrane conductance and *total* volume of distribution are the same for the permeant tracer as the corresponding values for the extracellular tracer. With these parameter values, do you expect that the curves for the extracellular and permeant tracers will look the same? Run the model plotting the outflow concentrations for the extracellular and permeant tracers over 30 seconds. Did the curves behave as you predicted? Why or why not?

EXPERIMENT EC-2

Change PS_g for the permeant tracer to 0.25 and PS_{ecl} to 0.75. The total conductance for the permeant tracer remains the same as in Experiment EC-1. How

will the outflow curve for the permeant tracer change from Experiment EC-1? How does this curve compare now to the extracellular tracer outflow curve? Why? (HINT: What is the ratio of *PS*/*V* for the extracellular tracer in Experiment EC-1? What are the *PS*/*V* ratios for the ISF and endothelial cell in EC-1? What are these ratios in Experiment EC-2?)

EXPERIMENT EC-3

Consider another situation that has the same conductance for the permeant tracer. This time allow the tracer to enter the interstitium through the endothelial cell as well as through the interendothelial clefts. Return to the starting values for parameter file 9endoth.par, and set the *PS* products for the permeant tracer to the following values:

Parameter	Value
PS_{ecl}	0.667
PS_{eca}	0.667
PS_{g}	0.25

How do the curves obtained in this experiment compare to those obtained in Experiment EC-2? Is this what you would have predicted? Why are the results different? (HINT: Remember that tracer can now enter the ISF from the capillary and from the endothelial cell.)

EXPERIMENT EC-4

The curves you have been examining in Experiments EC-1, EC-2, and EC- 3 do not necessarily look the same, nor do the tracers take the same amount of time to clear the exchange unit. Should the mean transit times of the extracellular and permeant tracers be the same or different in these experiments? Should the mean transit times of the permeant tracer change when you change the distribution of the apparent volume of distribution as you did in the previous exercise? Look at the mean transit times for the two tracers (Report window). Be sure to plot the data for sufficient time to obtain essentially the whole outflow curve. Are they the same or different? Is this consistent with what you predicted?

Summary

In the experiments of this exercise, you have seen that the effect of the endothelial cell on the shape of the output curves depends on the *PS* products. It is easier to predict the results when the two regions operate in parallel (Experiments EC-1 and EC-2) than when they are arranged as in real tissues where PS_g provides a “shunt” from the capillary to the ISF.

While the effect on the shape of the curves may be difficult to predict in detail, the effects on the mean transit time can be predicted exactly knowing only the flow and total volume.

2.3.5. *Selecting an appropriate input function*

Introduction

Choosing which input function to use depends, to some extent, on the information you are trying to derive from the model and the characteristics of the tracer you are using. When examining the characteristics of the model to learn more

about model behavior, the choice of input function may be somewhat arbitrary. When using the model to analyze experimental data, however, choosing an appropriate input function can be critical. Consider the following exercise.

Exercise: Choosing an input function (Parameter file 10expinput.par)

We are designing an experiment to examine the relative permeability-surface area products of two tracers that remain extracellular (e.g., inulin and sucrose). Both tracers have similar volumes of distribution within the interstitium, and neither undergo significant axial diffusion in the plasma or interstitium. We want to use the model to help decide if a bolus injection or a constant infusion of tracer will yield results that will allow us to distinguish between the two tracers. We must, of course, consider our measurement techniques. The relevant parameter values are summarized in the table below. Parameter file 10expinput.par has been initialized to use the inulin values for the extracellular tracer and the sucrose values for the permeant tracer. Again, one pathway is used, and the nonexchanging vessels are turned off.

	PS_g	D_p	V'_{isf}	D_{isf}
Inulin	0.16	0.0	0.155	0.0
Sucrose	0.5	0.0	0.16	0.0

This file has the input parameters for a constant infusion and for a LNDC input initialized as shown below. You can change between these two by clicking on the ‘Function’ button in the Vascular input window and selecting from the pop-up menu either the LNDC or pulse #1 function.

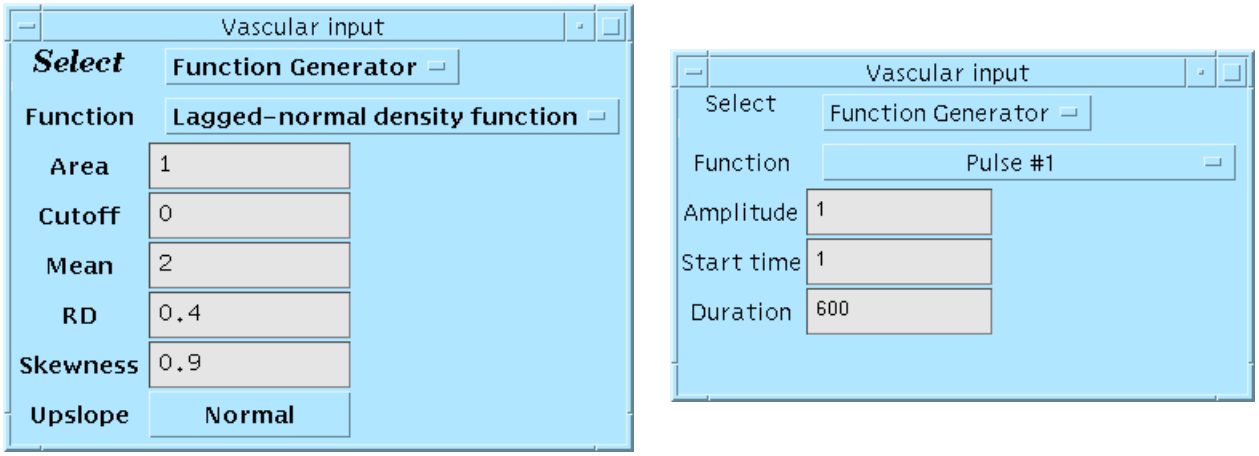


Figure 2.17. Vascular input options for exercise 10: LNDC (left) or constant infusion (right).

EXPERIMENT IF-1 In this case, you will observe (measure) the outflow concentrations of the two tracers. First, use a lagged normal density function as the input function, and then use a constant infusion of tracer as the input function. Compare the outflow concentrations of the two tracers to the outflow concentration of the intra-

vascular tracer. Do you think one type of input function is preferable to the other? (In answering this question consider the differences between the outflow curves. Which type of input produced the larger difference?)

EXPERIMENT IF-2

In this case, your conclusions will be based on the residue function of the tracers (i.e., the amount of tracer in the tissue). Change the plotting parameters to plot the amount of extracellular (*Q_e*) and permeant tracers (*Q_p*) in the tissue. Repeat the experiment, and examine an LNDC injection and constant infusion. How does the choice of tracer administration affect your ability to obtain meaningful data if you measure the residue function?

Summary

In the design of an experiment, the form of the tracer injection can significantly change the shape of the output function. These changes depend on the exchange parameters of the tracer being used, and may increase, or decrease, the accuracy with which specific parameters can be estimated from the data. The model that will be used for data analysis is a vehicle for examining these effects.

2.3.6. *Summary*

These exercises have focused on the basic indicator dilution principles of mean transit time ($t = V/F$ seconds) and membrane conductance. The mean transit time can be calculated from the outflow curve of any tracer that is completely recovered (i.e., not consumed and data collected for sufficient time) when the mean of its input is known. This calculation is valid even when the tracer crosses diffusible membranes; thus tracers that access different total volumes will have different transit times.

The conductance of the capillary membrane can be measured by calculating the extraction of a diffusible tracer compared to a reference (intravascular) tracer using the Crone equation: $PS = -F \cdot \ln(1 - E)$. This relationship is valid early in the outflow curve when backflux is negligible. The relationship between flow and *PS* determines whether the dynamics of the tracer are primarily dominated by flow (*PS*/*F* large) or by membrane transport (*PS*/*F* small).

The behavior of a specific tracer in an organ, controlled by the flow, membrane conductances, and volumes of distribution, often dictate that a given form of input function leads to better estimation of the system parameters. This decision is influenced by the type of data that can be collected. The model gives a tool for making this decision.

2.4. Advanced Exercises

2.4.1. *Introduction*

In this section, you will look at three factors that complicate the interpretation of indicator dilution curves: consumption of tracer, heterogeneity of flow, and the presence of nonexchanging vessels.

2.4.2. *The effect of tracer consumption*

Exercise: Effect of tracer consumption (Parameter file 11consump.par)

The primary assumption underlying indicator dilution techniques is that mass is conserved. That is, whatever mass enters the system leaves the system in the same form. If the chosen tracer undergoes transformation (e.g., the tracer is consumed) or leaves the system by a route other than venous outflow, this assumption is not valid.

Parameter file 11consump.par is set up to use a LNDC input for the permeant tracer and to plot the permeant tracer outflow curve. The tracer parameters are given below. (Remember that *Fp* is on the MMID4 model layout diagram, while the other parameters are on the permeant tracer BTEX unit diagram).

Parameter	Value
F_p	1.0
Permeant Parameters:	
V_p	0.01
PS_{ecl}	1.0
PS_{eca}	1.0
V'_{ec}	0.02
PS_g	1.0
V'_{isf}	0.15
PS_{pc1}	1.0
V'_{pc}	0.6

All consumption parameters (G) and diffusion coefficients (D) are set to 0.0. As with previous exercises, only one flow path is used and the nonexchanging vessels are omitted.

EXPERIMENT G-1 Load the parameters from parameter file 11consump.par and run the model. The plot shows the permeant tracer outflow when there is no consumption. Now let the tracer be consumed in the endothelial cell, i.e., set the value of G_{ec} to 2.0 ml $\cdot$ gm^{-1} $\cdot$ min^{-1}. Run the model again. How does this curve compare with the previous during the upslope? What about at the peak? Do the tails differ? Is this what you would have predicted? If consumption occurs, does the

transit time calculation still hold? If you know that consumption occurs, what steps could you take experimentally to ensure a reasonable measurement for the mean transit time? When performing experiments with consumable tracers, how important is it to include additional reference tracers? Why?

EXPERIMENT G-2

Repeat Experiment G-1 but this time examine the influence of consumption in the parenchymal cells. Reset consumption in the endothelial cell (*Gec*) to 0.0, and set the consumption in the parenchymal cell (*Gpc*) to 2.0. Will consumption in the parenchymal cells affect the outflow curve to the same extent as consumption in the endothelial cells? Why? Run the model again to confirm your prediction.

You can use the XSIM loops to compare the curves while keeping them on the screen. Reset G_{pc} to 0.0. Set the number of iterations for both the inner loop and the outer loop to 2 (Loop controls are located under *Model: Run config...*).

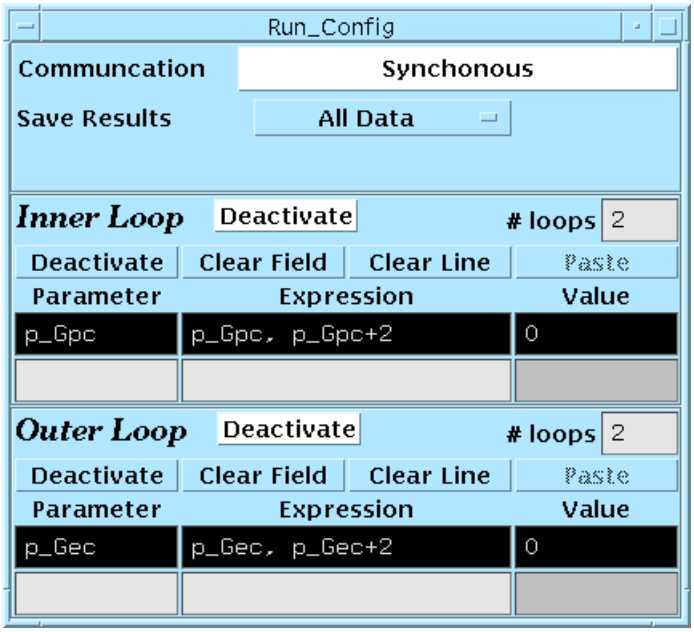


Figure 2.18. The Run Config window configured for Experiment G-2.

These loops have been preset to modify the consumptions. Running the model will produce four curves: (1) G_{pc} = 0.0 and G_{ec} = 0.0, (2) G_{pc} = 2.0 and G_{ec} = 0.0, (3) G_{pc} = 0.0 and G_{ec} = 2.0, and (4) G_{pc} = 2.0 and G_{ec} = 2.0. Run the model and observe the increasing effect of consumption. This linear scale best shows the effect on the peak. Use a semilog plot to show the effect on the tails better. When XSIM creates a semi-log plot, it will default to a linear plot if the y-axis minimum is less than or equal to zero, so you will need to either turn on the y-axis autoscaling, or enter a y-axis minimum that is greater than zero.

EXPERIMENT G-3

When a tracer is transformed, it may: (a) be removed by the system or (b) stay within the tissue in the transformed state. When examining the residue function (i.e., the amount of tracer in the tissue, as in PET studies), it is important to rec-

ognize the difference between these possibilities. Reload the parameters from parameter file 11consump.par. Change the plotted y parameter to be the residue curve for the permeant tracer in the tissue, *Q_p*. Run the model and observe the curve. This is the content of the tissue when there is no consumption. Turn on consumption in the endothelial cell again (i.e., set *Gec* to 2.0). Should this cause the residue curve to increase or decrease? Why? Run the model. Was your prediction correct?

Now plot Q_p for the exchange units (*Q_cap_p*). Since all the nonexchanging vessels are turned off, all the tracer in the tissue is in the exchange units. Run the model. Are the curves the same? Why not? The answer to the latter question has to do with the way in which the two curves are calculated. The residue curve stored in parameter *Q_p* is calculated from the integral of $C(t)_{in} - C(t)_{out}$. Thus all tracer that does not appear in the outflow is included in the residue; this is equivalent to case (b) above. The residue curve stored in parameter *Q_cap_p* is calculated from the summation of the tracer concentrations in all segments of all regions in the BTEX unit. Since tracer that is consumed no longer affects the regional concentrations, this is equivalent to case (a).

In a PET experiment in which the tracer in the tissue is being monitored externally, what must you know about tracer kinetics to interpret the obtained data correctly? The MMID4 Reference section of this manual gives more details about the various residue curves that are calculated by the model.

Summary

When some of the tracer is consumed in its transit through the system, the mean of its outflow curve no longer reflects the theoretical mean transit time of the system. Consumption of tracer in endothelial cells usually has a greater effect on the outflow curve than parenchymal cell consumption because these cells have more immediate access to tracer entering in the plasma.

MMID4 uses two methods to calculate residue curves, integration of outflow curves and summation of content calculated in individual regions. The nature of the measurement being made determines which measure is the appropriate one.

2.4.3. *The effect of flow heterogeneity*

Exercise: Heterogeneity of flow (Parameter file 12hetero.par)

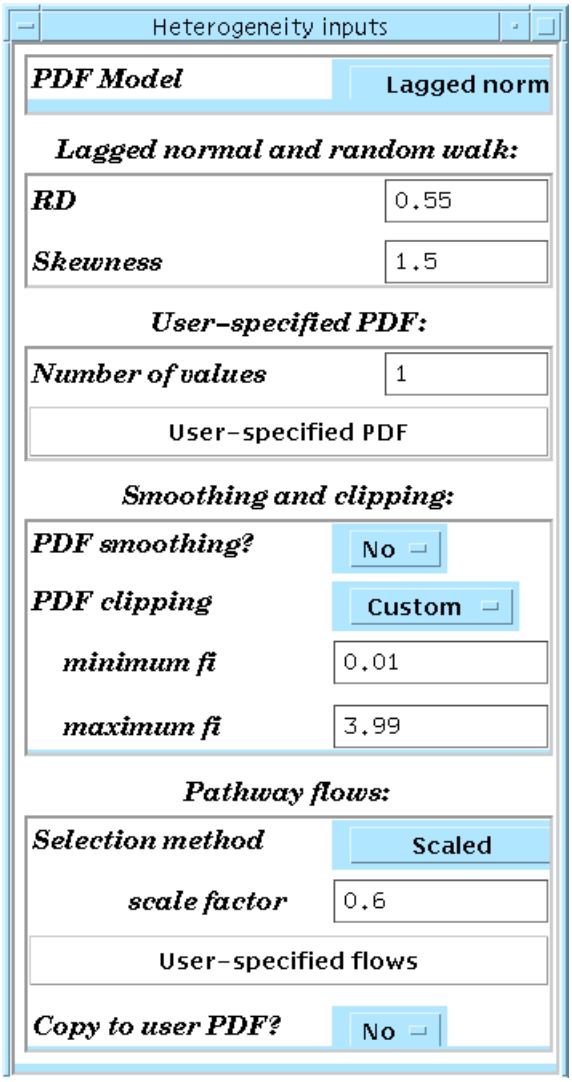
Thus far you have focused on exchange occurring in one pathway. It is reasonable to assume, however, that in all tissues there is some degree of flow heterogeneity. You must, therefore, be concerned with how flow heterogeneity affects the outflow and residue data that we collect in tracer experiments. Parameter file 12hetero.par has been initialized with parameters for this exercise. Load the values and check the values of the transport parameters for the three tracers. Note that the parameters that are common to two tracers have been set to the same values (e.g., PS_g for the extracellular tracer, and PS_g for the permeant tracer, are both set to 1.0).

The plotting parameters are initialized to plot the intravascular tracer outflow curve in plot area 1 and the flow probability density function (PDF) in

plot area 2. Run the model and observe the curves. Note that since only one pathway is being used, the flow PDF appears as a square wave.

EXPERIMENT H-1

Begin this study by comparing outflow curves when one pathway is used with those obtained when 20 pathways are used. The parameters that define the flow heterogeneity can be observed by opening the Heterogeneity inputs window, found at *Parameters: Flow Heterogeneity > Heterogeneity inputs*.



Use a LNDC model for the shape of the flow heterogeneity.

Set the *RD* of the heterogeneity.

Set the skewness of the heterogeneity.

Section is not used in this example, since the PDF is model-specified.

Use user-defined minimum and maximum flow limits.

Set the minimum relative flow.

Set the maximum relative flow.

Set the model for a ratio of time to flow domain spacing.

Set the model ratio to favor time domain spacing.

User-specified flows not used in this example.

Figure 2.19. Heterogeneity inputs configured for the Experiment H-1 lagged normal density curve.

Turn on the loops by setting the number of inner loop iterations to 2. The inner loop has been set to increment Npath_int, the number of paths, by 19. Run the model and observe the intravascular outflow and flow PDF at 1 and 20 paths. Why are the two outflow curves different? Do the two curves have the same mean transit time?

Note that the second solution, 20 pathways, took longer than the first. The model had to evaluate 20 BTEX operators instead of just one. The solution didn't take twenty times as long because there is a constant computation overhead for calculation of curve statistics, plotting, etc. Also, the axes on the flow PDF plot seem to run from 0.0 to 10.0. This is because XSIM always uses the limits of the independent variable for x-axis scaling. In MMID4, the actual scale of flow PDF plots starts at 0.0 and runs to the value of *Plotmax_h* (*Parameters: Flow heterogeneity> Flow heterogeneity results ^Plot maximum*), which is set to 4.0 in this parameter file.

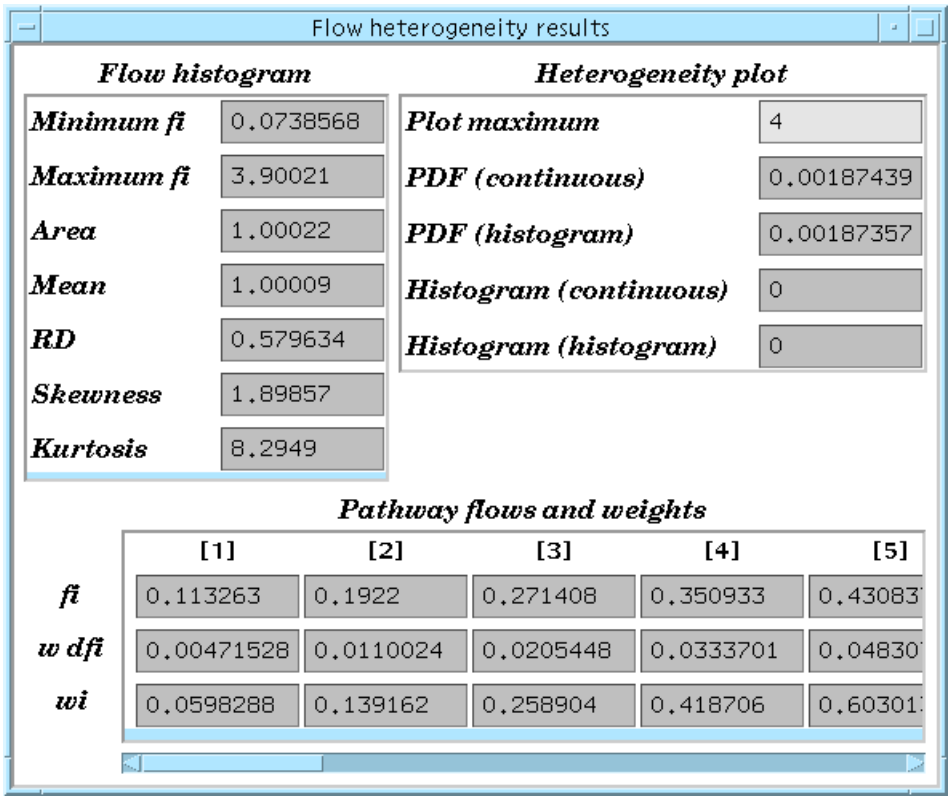


Figure 2.20. Flow heterogeneity results. Note the Plot maximum input field at upper right.

Change the output parameters so that the outflow of the extracellular (*Cout_e*) and permeant (*Cout_p*) tracers are plotted in plot area 1 instead of the intravascular outflow. Considering the changes that you saw in the intravascular outflow, what changes do you expect in the other tracers? Run the model. Are your predictions correct?

Now look at the effect on the tail of the permeant curve. Turn off the plotting of the extracellular curve and make the following additional parameter changes: Set the ending time to 60.0 (*Parameters: time*), the plot style to semi-log plot, and manual scaling with a y-minimum of 1.0e-4 in plot area 1. Set the plot period so that every 10th point is plotted. In the *Model: Run Config* window change the Save Results button to Every Nth datum, and enter 10 in the value field that appears.

What changes do you expect? Why? Run the model and check your prediction.

XSIM Note: The plot period controls how often the solution points are plotted and saved. If N is 1, the solution at each time step is plotted; if set to 10, every tenth point is plotted. For some models with many output parameters, it may be useful to increase the plot period to avoid using up the available computer memory. Since there is only a small time overhead in plotting, increasing the plot period will not significantly speed up a solution. It should be noted that the accuracy of the calculations is not effected as increasing the time step might.

Finally, change the value of PS_{pc1} from 0.1 to 1.0 and rerun the model. How does this increase change the outflow curves for the single and multi-path solutions? Vary other permeant tracer parameters. Can you predict how these changes will affect the solutions?

Summary Flow heterogeneity alters the shape of the tracer outflow curves. This is due to the presence of high flow pathways that affect the leading edge and peak of the curve and low flow pathways that affect the tail. The magnitude of these changes are dependent on the transport parameters of the organ and the parameters of the flow distribution.

EXPERIMENT H-2 How much flow heterogeneity is necessary before changes can be seen? For this experiment, we will use another mode of defining the heterogeneity. Reload the starting values of parameter file 12hetero.par, and edit the flow heterogeneity parameters (*Parameters: Flow Heterogeneity >Heterogeneity inputs*). Change the *PDF model* setting to User-specified PDF; this specifies that you will enter a flow distribution instead of using a mathematical model like LNDC. While you are here, also change the Number of values to 2 for the User-specified PDF, and the Selection method under Pathway flows to *Equal F spacing*. Examine the user-specified flow distribution by clicking on the User-specified PDF button.

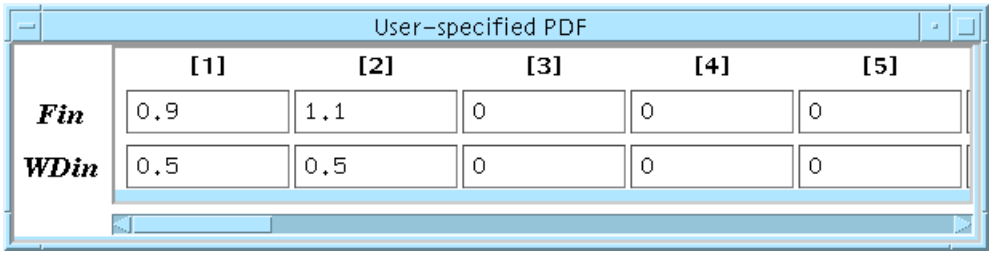


Figure 2.21. User-specified PDF with 2 flow classes designated.

The window that should open (Fig. 2.21) shows a distribution with 2 classes having flows of 0.9 and 1.1 with equal weights of 0.5 and 0.5. How will this small degree of heterogeneity change the outflow for the vascular tracer? Before you run the model to see the answer, open the Run Config window (under *Model* menu), change the number of inner loop iterations to 2 and change the increment of Npath_int to 1.

You can increase the amount of heterogeneity by decreasing *Fin[1]* and increasing *Fin[2]*. Keep them symmetrical around 1.0. Try the values of 0.75 and 1.25 for the flow. How much difference is seen? Now try 0.5 and 1.5.

Summary Flow heterogeneity may be present but not have a significant effect on the outflow curves depending on its magnitude. In real organs, however, the level of heterogeneity is such that it should nearly always be considered.

2.4.4. *The influence of nonexchanging vessels*

Exercise: The influence of nonexchanging vessels (Parameter file 13nonex.par)

After a tracer is injected into an *in vivo* or *in perfuso* system, delay and dispersion of the indicator occurs within the large vessels between the injection site and the exchange unit. Similarly, delay and dispersion occurs on the venous side between the exchange unit and the sampling site of the outflow curves. The relative dispersion that occurs in the arterial component is about 18%. The degree of dispersion in the venous vessels is not known but is probably similar to or somewhat greater than that seen in the arteries. To account for this delay and dispersion, MMID4 includes several nonexchanging operators on the arterial and venous sides (see Fig. 1.1). The parameters defining a nonexchanging operator are its volume, expressed as ml/g of organ, and its relative dispersion, *RD* (Fig. 2.23). The volume of the nonexchanging vessels does not contribute to the plasma volume in the exchange units (*Vp*).

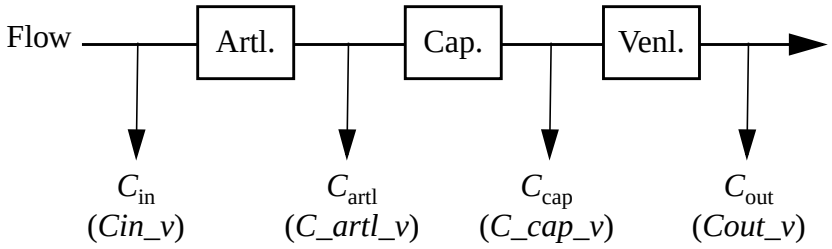


Figure 2.22. Sampling sites for effects of nonexchanging vessels exercise.

EXPERIMENT As an introduction to the effects of nonexchanging vessels on the outflow curves from the organ, load the parameters in parameter file 13nonex.par. Run the model and note that two curves, the intravascular tracer input and outflow, are plotted. In fact there are four curves being plotted. In addition to C_{in} and C_{out}, the outflow from the arteriole and from the capillary are plotted. Look at the parameters for the nonexchanging vessels found on the MMID4 model layout diagram. Do these values explain why you only seem to see two curves? (HINT: When the volume of a nonexchanging operator is 0.0, it is effectively removed from the solution.) Set the volume of the arteriole, to 0.02 ml/g and run another solution. Then set the volume of the venule to 0.03 and get a new solution. Note that the arteriole and venule are dispersive, *RD* of 18% and 20% respectively, but that no dispersion occurs in the capillary. Why not? Try changing D_p (*Parameters: vascular/microvascular operators > tissue-capillary units > BTEXunit (vascular tracer) ^Dp*) to 0.00015 and running the

model. How else could you make the capillary dispersive for the vascular tracer?

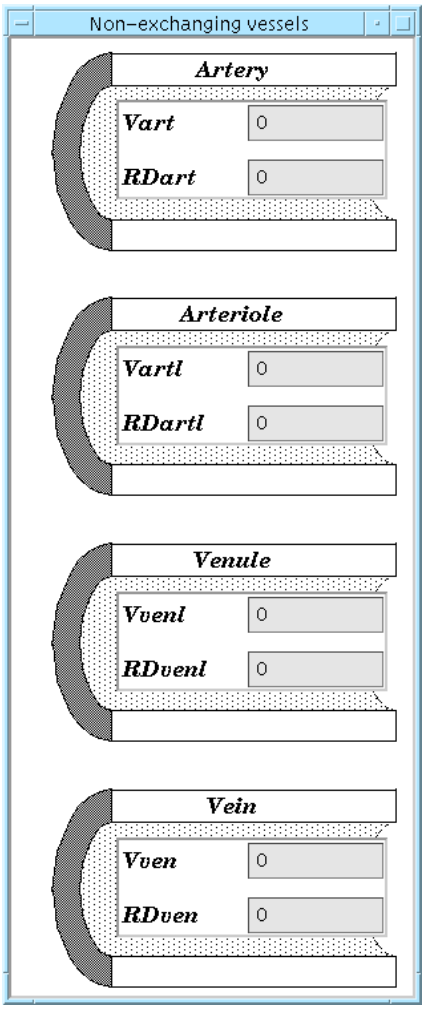


Figure 2.23. Non-exchanging vessels window.

Summary

In MMID4, a nonexchanging vessel is characterized by its volume and *RD*. No tracer leaves the nonexchanging vessels by diffusion or by consumption. Their effect is by delay and dispersion.

2.4.5. *Summary*

The interpretation of outflow dilution data is complicated by factors such as consumption, heterogeneity, and diffusion in nonexchanging vessels. Exercises introduced you to these concepts and to ways that the MMID4 model can be used to examine these factors and correct for them in future modeling analysis.

2.5. Analysis Exercises

2.5.1. *Introduction*

Thus far, you have used MMID4 in a predictive mode. That is, you have assumed that the model provides an accurate description of exchange within the capillary-tissue unit (within certain limits) and have used the model to predict what you would see, either within the exchange unit or at the outflow, if the various parameters took on certain values. In this mode, you can run a number of "experiments" with the model to predict what might happen under similar conditions in a living system. To test the model's validity, you could run those experiments in a living system and compare the actual data to the model's predictions. If the predictions match the actual data, you would assume that the hypotheses used in building the model are reasonable and that the model provides one explanation for the mechanisms involved in the exchange process.

You can also use the model in this mode to help design experiments that will be run in a living system. Predictions with the model may help decide what initial tracer concentrations to use, how the tracer should be administered (e.g., bolus or continuous infusion), what sampling times are optimal, or, perhaps, which sampling site will yield the most meaningful data.

The model, however, can also be used in an analysis mode. Once the model's validity has been established under a variety of conditions, you can assume that the model can be used to explain phenomena occurring under the same or similar conditions. If this is the case, the model can be used with data from real experiments involving multiple tracers to determine the values of model parameters during the conditions of the experiment. This is another way of testing the model. If the resulting parameter values are not consistent with what is already known or if these values are not internally consistent, the hypotheses upon which the model is based do not adequately explain the phenomenon being examined, and the model must be revised. In this analysis mode, the data from the real experiment serves as a reference for the model output. The model is run with a set of parameter values, and the output is compared to the reference data. The parameter values are adjusted, and the output is again compared to the reference data. This process continues until the model output and the reference curves are essentially the same. This process, called parameter optimization, assumes that the reference data accurately reflect what is happening in the system (i.e., the data are of high quality, and the experimental design has yielded appropriate data). This is a critical point because a number of factors can cause significant errors to be introduced into the experimental data.

The exercises in this section focus on parameter optimization and on using the process of deconvolution to obtain an input function from the outflow curve of an intravascular tracer. Both these processes require that you provide the model with a reference data file. Reference data for use with XSIM reside in

files with a TAC (Time Activity Curve) format and were copied into your directory when you copied the tutorial database.

2.5.2. *Parameter optimization*

XSIM provides an automatic parameter optimization feature. However, to best understand the process, it is helpful to go through the process manually.

For this exercise, the reference data file has been generated using the model. Hence, the assumption that the reference data is error-free is valid. A parameter file has also been set up to serve as your starting point for optimization. In this parameter file, the input function, total flow and distribution of flow used to generate the reference data have been retained.

Exercise: Manual parameter optimization (Parameter file 14manopt.par)

Introduction

In this exercise, your task is to adjust the appropriate parameter(s) of the model so that the model solution fits the data in the reference file for the outflow of the intravascular, extracellular, and permeant tracers. For this exercise, you can assume that:

1. the input function is known;
2. the flow is known;
3. the distribution of flow is known;
4. PS_g and V'_{isf} are the same for the extracellular and permeant tracers;
5. the permeant tracer does not enter endothelial cells;
6. there is no axial diffusion of any tracer; and
7. there is no consumption of any tracer.

Begin the exercise by loading parameter file 14manopt.par. A window will appear asking if the reference file “pf14.tac” should be loaded. Press the Continue button to do so.

The intravascular tracer

Run the model. Two curves appear. The first, shown by a series of solid circles is the outflow concentration of the intravascular tracer from the reference file. The curve plotted as small open circles is the model solution for the intravascular tracer outflow with the current parameter values.

If the mean flow and the flow distribution are “correct” as set in the parameter file, what other parameters will influence the outflow curve of the intravascular tracer?

Adjust the plasma volume (*Vp*) until the model curve matches the reference curve. Note that plotting the curve on linear axes will enable you to view the goodness of fit at the beginning of the curve, while using a semilog plot will allow you to examine the fit at the tail of the curve. However, you will notice that XSIM maintains a linear plot when a logarithmic y-axis is chosen, since the reference curve contains data points equal to zero. Change to manual scaling, and enter a small, but nonzero, y-axis minimum, such as 1.0e-12.

Fitting the intravascular tracer output curve to the intravascular tracer reference data has provided an estimate of the plasma volume. The next step is to use this information to focus on the extracellular tracer.

The extracellular tracer

To fit the extracellular data you need to display the extracellular tracer reference data and model output on the plot. The extracellular data are the second curve in the data file. In the plot area window, click with the right hand mouse button in the field under Reference (this should contain the parameter 'R1s1'). In the window that appears, click on the second line, labeled 'R1s2'. Change the plotted parameter from *Cout_v* to *Cout_e* to display the model solution that corresponds to this data curve. You should also start with a linear plot.

Adjust PS_g and V'_{isf} for the extracellular tracer until you have achieved a good fit to the reference curve. First, try decreasing PS_g. What part of the curve is altered? Does it get closer to the desired curve or further away? Keep working with PS_g until you have fit that part of the curve. Now shift to a semi-log plot and modify V'_{isf}.

The permeant tracer

In accord with assumption 5 above, insure that permeant tracer parameters PS_{ecl} and PS_{eca} are set to zero. Set the plot parameters to show the permeant tracer curves by changing the plotted reference curve to 'R1s3' and the plotted model parameter to *Cout_p*. This parameter file has been set up so that the PS_g and V'_{isf} for the permeant tracer always have the same values as the corresponding parameters for the extracellular tracer (assumption 4). To see how this is done, double click on the *PSg* button and note that the field Eval is filled with the expression *e_PSg*. You can only change permeant *PSg* by changing the extracellular *PSg* value, unless you erase the permeant Eval field.

Repeat the optimization process adjusting PS_{pc1}, and V'_{pc} until a reasonable fit of the model output to the reference data is achieved. Since the parenchymal cell is "deeper" than the ISF, the parenchymal cell parameters affect the output later than the ISF parameters. Try decreasing PS_{pc1}. What part of the curve changes? Does changing V'_{pc} have a larger effect? On which part of the curve?

Summary

You have now completed the fitting of the three data curves. Several assumptions were made in this exercise. Which of these do you think are correct? If the extracellular and permeant tracers are of different molecular weights and/or have different charge characteristics would they both permeate the gaps between the endothelial cells in the same way? If the permeant tracer crosses the membrane of the parenchymal cell would it also permeate the endothelial cell?

The values for the parameters used to generate the reference curves are shown in Table 2.4. How close to these values are your final values?

A strategy to use for modifying parameter values can be based on seeing which part of the curve is affected by each of the parameters and the changing the parameters so as to fit different sections of the curve in turn. When only two parameters are involved, it may be feasible to do this manually. Even with two parameters, however, there is interaction, and with three or more parameters the task quickly becomes unmanageable.

Table 2.4: Parameter values used to generate curves in pf14.tac

Tracer	Parameter	Value
Intravascular	V_p	0.05
Extracellular	PS_g	1.00
	V'_{isf}	0.15
Permeant	PS_g	1.00
	V'_{isf}	0.15
	PS_{pc1}	1.20
	V'_{pc}	0.80

Exercise: Automated parameter optimization (Parameter file 15autoopt.par)

Introduction

XSIM includes an automated parameter optimizer. To demonstrate its use, you will use the optimizer to obtain fits to the same three curves that you used in the last exercise. You will use the same parameter file and make the same assumptions regarding the model and tracers. At the end of the exercise, you will check your results using parameter file 15autoopt.par.

In general, the optimizer used by XSIM uses the same approach that you used in your manual optimization. It optimizes the fit of one curve at a time. In deciding how to modify parameters, it looks to see which part of the solution is sensitive to each parameter being optimized and modifies the value accordingly. This "sensitivity optimizer" (SENSOP) is described in detail by Bassingthwaighte, Chan, and Goldstein (1993).

To set up XSIM to optimize parameter values you must specify the following:

1. the name of the reference data file;
2. the indices of the reference data curve and the parameter variables of the simulation outputs that are to be compared;
3. the weighting scheme that is to be used;
4. the names of the parameters that are to be optimized and the limits to be placed on them; and
5. the optimizer stopping criteria (i.e., the limits used to determine when a "good" fit has been achieved).

XSIM Note: Details of automated optimization under XSIM are given in the Interface Reference Manual Section 8.2.4.

This exercise does not cover all the details of parameter optimization under XSIM. Items 1, 3, and 5 have been preset in the parameter file. The instructions below only deal with items 2, and 4.

Begin the exercise by recalling the original values in parameter file 14manopt.par. Run the model. You should see the same two curves that you saw at the start of the last exercise.

Intravascular tracer

The initial parameter values have been set for optimization of the plasma volume using the data for the vascular tracer. View the BTEX unit vascular parameters. The value of the plasma volume is set to 0.02. This value was used to produce the curves shown and will be the starting value used by the optimizer. Select *Optimization Graph* from the *Results* menu and *Optimize Config...* from the *Model* menu, then start the optimizer by clicking on the optimize button in the main window.

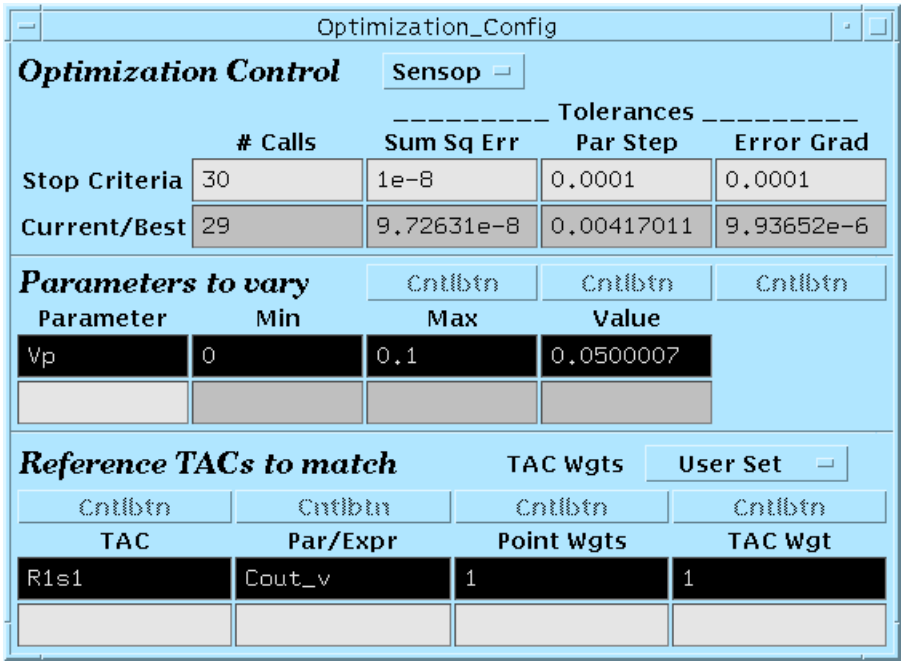


Figure 2.24. The Optimization Config window after optimizing Vp.

The updated parameter value is displayed under the 'Parameters to vary' section of the Optimization Config window. Note how the value changes as the optimizer moves toward the best fit. Under Tolerances is shown the value of the sum of the squared errors. This number will decrease as the fit gets better. Run a solution. Note the new value of plasma volume. How well does this compare to the value in Table 2.4?

(NOTE: After running an optimization, you should *always* run the model and see how well the solution fits the reference data. Though appropriate stopping criteria may be set and the variation between the solution and data minimized, visual inspection will reveal any systematic deviations in the solution. Such systematic biases may indicate that the model being used is not appropriate for the data being analyzed. The bias may be overcome by using a feature of the model that is not currently being used. In this exercise, for example, the

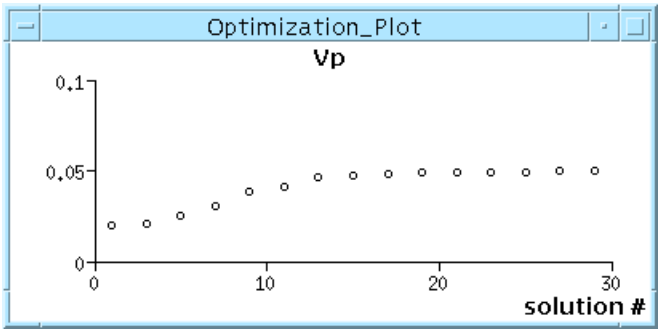


Figure 2.25. The Optimization Graph window.

nonexchanging vessels are not being used. Obtaining a good fit to some data may require their usage.)

Table 2.5: Parameter values for optimizing the extracellular tracer parameters

Usage	Value
Plot Area 1 window:	
Plot Reference curve	R1s2
Plot Y Parameter	Cout_e
Optimization Config window:	
Optimize parameter	e_PSg
PSg maximum value	2
Optimize parameter	e_Visfp
Visfp maximum value	1
TAC data curve to be matched	R1s2
Solution curve	Cout_e

Extracellular tracer

The next step is to modify the parameter values so that you can optimize the extracellular tracer parameters. (The changes required are summarized in Table 2.5.) First change the plot parameters for the reference data (R1s2) and the simulation output (Cout_e) so that the extracellular tracer curves are shown on the plot.

You can change the remaining parameters by using the Optimization Config window shown in Fig. 2.24 (*Model: Optimize Config...*). Enter each parameter name in the Parameter field either by typing, selecting from the pick control window (right mouse button), or by using the paste button. The minimum, maximum and current value of each parameter is filled in by XSIM. Change the maximum value of *e_PSg* to 2.0 by editing the field directly. Change the reference TAC curve to R1s2 and the matching curve to Cout_e.

By making these changes you have directed XSIM to optimize the value of two parameters, PS_g and parameter V'_{isf}. PS_g can have a minimum value of 0.0 and a maximum value of 2.0; V'_{isf} can vary between 0.0 and 1.0.

XSIM Note: The minimum and maximum values set for a parameter being optimized are also used as the y-axis scale for the plot of parameter value versus model iteration during optimization.

You have also told XSIM that the reference data are found in data set 2 of the reference data file and that the model solution that is to be fit to the data is Cout_e.

XSIM Note: When optimizing parameters it is essential that the model output and the reference data curve are comparable. (In the exercise above, for example, both are composed of the outflow curve for the extracellular data.) If this is not the case, XSIM will still do its best to get the best fit of the model to the data, but the results will be meaningless.

Note the starting values of PS_g and V'_{isf} in the Optimization Config window. Run a solution and then start the optimizer. This time two plots are produced, one for PS_g and one for V'_{isf}. When the optimization is complete, run a solution and check the optimized values of PS_g and V'_{isf} against those in Table 2.4. Are they close?

Table 2.6: Parameter values for optimizing the permeant tracer parameters

Usage	Value
Plot Area 1 window:	
Plot Reference curve	R1s3
Y axis Plot Style	log
Plot Parameter	Cout_p
Optimization Config window:	
Optimize Parameter	p_PSpc1
p_PSpc1 upper limit	2
Optimize Parameter	p_Vpcp
p_Vpcp upper limit	1
Solution curve parameter	213
Data curve parameter	R1s3

The permeant tracer Finally use the permeant data to get values for PS_{pc1} and V'_{pc}. The parameter changes that you need to make are shown in Table 2.4. Note that you will use a semilog plot this time to emphasize the fit to the tail of the curve.

When the parameter changes are made, run the optimizer, then the model, and compare the results for PS_{pc1} and V'_{pc} with Table 2.5. How closely do the optimized values agree with those you obtained by your manual optimization?

Summary

The automated parameter optimizer obtains the best fit of up to four solution curves to the corresponding data curves. Up to eight parameters can be optimized during each optimization run. In this exercise, the starting values of the parameters and the optimizer stopping criteria have been chosen such that the optimizer will stop after relatively few model iterations. You can test how the starting values affect the optimization process by recalling the original values of the parameter file, choosing starting values that are further away from the "true" values and repeating the exercise.

If you optimize the curves one at a time, as done above, the order in which you do the optimizations is important. The permeant tracer analysis depends on the answers obtained from the extracellular tracer which in turn depends on the vascular tracer answer. Since XSIM can optimize eight parameters and three curves at once, you could get the values of V_p, PS_g, V'_{isf}, PS_{pc1}, and V'_{pc} in a single run. Parameter file 15autoopt.par is set up to do this optimization. To see the results, load the parameters, open the Optimization config and Optimization graph windows, run the optimization, and check the values. This optimization run will take some time, but at the end you will see that the results are quite good. Remember, however, that the reference data are exactly the model solution; they are noise-free. The presence of noise can significantly effect the ability of the optimizer to extract meaningful parameter values from the data. This is demonstrated in a later exercise.

2.5.3. *Using deconvolution to generate an input function*

Introduction

A critical assumption made in the optimization exercises was that the input function was known. What can be done if the input function is not known? One approach is to use outflow data in conjunction with a technique called deconvolution to generate an input curve. This technique starts with determining the response of the system to a specific input (an impulse function). This gives the system transfer function (see Fig. 2.26). The transfer function describes how the system behaves with any input function to yield its output function. If data are available that reflect how the system behaved in an experiment (i.e., an output curve), the transfer function can be used with these data to "back calculate" (deconvolve) and obtain an input function. Mathematically, the process is not as simple as it sounds, however. The output function used by MMID4 to obtain an input curve is the outflow curve for the intravascular tracer.

To obtain a reliable estimate of the input function using deconvolution, the model must be well defined for the intravascular tracer. This means knowing or have good estimates of total flow, plasma volume, and distribution of flow within the organ. Flow is generally obtained through direct measurement during the experiment. Plasma volume is either measured or, for some preparations, estimated from existing data in the literature that indicates that plasma volume is relatively constant. The distribution of flow is obtained by injecting

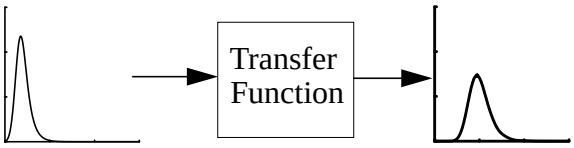


Figure 2.26. The transfer function is a mathematical description of the relationship between the input and output functions. For deconvolution, the response of the system to an impulse function is determined.

radiolabeled microspheres and determining their distribution either by external counting or by sectioning the tissue and counting it.

Exercise: Deconvolution (Parameter file 16decon1.par)

Load the values from parameter file 16decon1.par. This exercise uses noise free data. The reference data were created by using a lagged normal density curve (LNDC) as input to a 2 region model and saving the results in a reference data file.

Run the model. The vascular tracer input and output curves from the reference data (large filled symbols) and the model (small open symbols) are plotted. The parameters are set to interpolate the first reference data curve as the input function. This is the same input function that was used to generate the outflow data reference curve; thus, the model curves pass exactly through the data points.

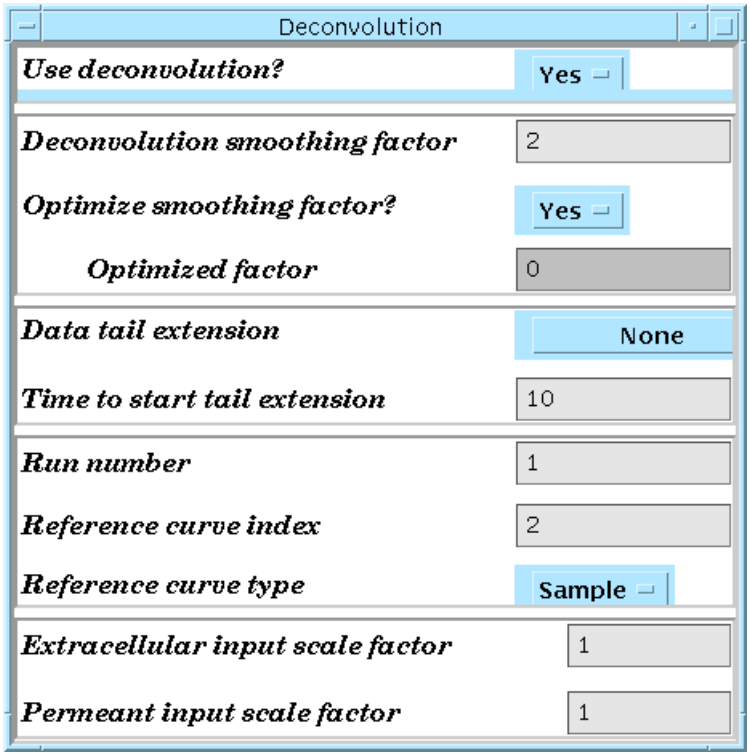


Figure 2.27. The Deconvolution window.

Now edit the deconvolution parameters (*Parameters: Input Functions> Deconvolution*). The deconvolution parameters are set to their default values. A detailed discussion of these parameters is in Section 3.2.3 and is not repeated here.

Specify that deconvolution is to be used to generate an input function by changing the Use deconvolution? button from No to Yes. Run the model again and observe the new input and output functions. How well do they fit the data? Does changing the values of the deconvolution smoothing parameters improve the fit? Why or why not?

Summary

Several important points should be remembered regarding the use of deconvolution. It is quite important that the reference data curve has high temporal resolution, covers an appropriate length of time, and have a low level of noise. Regarding temporal resolution, linear interpolation is used to predict the points at the simulation times that lie between the actual data points; thus it is desirable to have data sampling at about the same frequency as the simulation time step. The time span that is appropriate for the data depends upon the system being modeled. As a rule of thumb, it is recommended that the data span at least:

$$3 \cdot SD_{\text{output}} + \bar{t}_{\max}, \tag{2.4}$$

Where SD_{output} is the standard deviation of the output curve and $t_{\max}$ is the maximum mean transit time through the system. If, for example, the lowest flow pathway of the organ has a mean transit time of 15 seconds and the output function has a standard deviation of 12 seconds, the data should be collected for 51 seconds. The problem, of course, is that you don't know the maximum mean transit time or the standard deviation of the output curve when you are doing the experiment; these are quantities that will come from the data and the analysis. You need to make educated estimates of the transit time and standard deviation. It is advisable to overestimate them since it is easier to discard data that you have than to fabricate data that you don't.

It is also important that the model used for deconvolution be a reasonable representation of the real system. This pertains specifically to the flow distribution. For example, if the actual distribution of flows in the organ had a relative dispersion of 0.5 and a skewness of 1.5, this could not be well represented by a model that used only two flow paths. Trying to deconvolve with such a model would produce anomalous results that bear little resemblance to the desired data. (You can try this experiment by running the above exercise again and setting the number of paths (*parameters: MMID4 model layout ^Path)* to 2.) Again a rule of thumb: start with the maximum number of paths. If you are to make many simulation runs and need to reduce the execution time of the model (which increases linearly with the number of paths), only reduce the number of paths to a level where no effect is seen on the calculated input function.

Exercise: Deconvolution with nonexchanging vessels (Parameter file 17decon2.par)

In the previous exercises, only the exchange unit was considered. In the real system, however, the tracers must also traverse the nonexchanging vessels. Parameter file 17decon2.par contains parameters similar to those in file 16decon1.par except that arterioles and venules are also included. Load the parameter file and check the values set for volumes and dispersions of nonexchanging vessels (*Parameters: MMID4 model layout ^artery, arteriole, venule, vein*). How would you expect that the measured output would compare with that in the exercise above? Run the model and observe the measured output curve. (Note that the time scale now covers 30 seconds instead of 15.) The parameters do not specify an input function. You can terminate by pressing the Cancel button in the main window.

Repeat the deconvolution process by changing the Use deconvolution? button from no to yes and running the model. Compare the calculated output to the reference data. How well do the two curves match? Did including the small vessels affect the deconvolution process?

2.5.4. *Working with noisy data*

Introduction

An important feature of the reference data used in the exercises above is that they are noise-free. They are the output of the model that was used to produce them. In the real world, data are never noise free. Some of the sources of error in experimental data are listed in Table 2.7. The magnitude of these errors can often be reduced by good experimental design and careful laboratory procedure. They can never, however, be totally eliminated. The exercises below briefly examine the effect of noise on parameter optimization and deconvolution.

Table 2.7: Potential sources of noise in multiple indicator dilution experiments

Sampling:	• Physical loss of sample (e.g., by splashing)
	• Cross-contamination of sequential samples
Pipetting:	• Errors in pipetting
	• Errors in sample dilution
	• Selective absorbance of tracer on pipette tips or glassware
Counting:	• Errors in spillover and quench correction of tracer samples
	• Technical errors in liquid scintillation counting
Other:	• Errors in dose quantification
	• Chemical transformation of tracer species
	• Tracer contaminants

Exercise: Parameter optimization with noisy data (Parameter file 18noisopt.par)

Parameter file 18noisopt.par is set up to optimize the PS_g, V'_{isf}, PS_{pc1}, and V'_{pc} using the permeant tracer outflow curve as the reference data. In this respect it is similar to the exercise you did using parameter file 15autoopt.par. What makes it different is that it uses a reference data file to which random noise has been added. Load the parameter file and run the model. Note that the reference data are no longer a smooth curve, but are somewhat irregular. These data have had 10% noise added to them. Open the Optimize Config window so that you can see the values of the parameters being optimized. Run the optimization and, upon completion, run the model. How well does the solution pass through the data points? The correct parameter values are shown in Table 2.4. How well do the optimized values agree?

Return to the main menu and reload the parameter file 18noisopt.par. This insures that the starting values for the parameters being optimized are the same as they were above. Change the reference data curve to run 2 by setting both the reference TAC field (Optimization Config window) as well as the reference field in the Plot Area 1 window to R2s3. Run the model. The data shown have 20% noise. Run another optimization and display the solution. Does the solution still pass through the data? How close are the optimized parameters to the correct values now?

Return to the main menu and recover the original parameters once more. Select the third run by changing the optimization reference curve and the plotted curve to R3s3. Repeat the optimization process. How do the solution and parameters compare now? Which parameters are closest to their correct values? If you were watching the optimization plot carefully, you may have noted that the optimizer used all thirty evaluations of the model that were permitted. Could it get a better fit if it ran longer? If you start another optimization run at this point, the optimizer will use the current parameter values (i.e., those resulting from the last optimization) and can run another thirty iterations. Start another optimization run. How many iterations are used before the optimizer stops? Is the fit any better or the parameters any closer to the desired values? With the current stopping criteria, this is the best the optimizer can do. Reducing the stopping criteria values may result in a somewhat better fit, but the basic problem is the amount of noise in the data.

Exercise: Deconvolution with noisy data (Parameter file 19noisdec.par)

Parameter file 19noisdec.par is set up to do deconvolution on several curves with increasing noise in the reference data. Load the parameter file and run the model. The plot area 1 shows the data with 10% noise and the model solution using the deconvolved curve as input. The plot area 2 shows the fit to the noise-free input and output curves. Is the fit to the output curve much different than you obtained with noise-free data? (You may want to review the results of the deconvolution exercise above.) How different is the fit to the input curve?

Change to the third reference curve by setting the reference curve index to 3 in the Deconvolution window (*Parameters: Input functions > Deconvolution*), deactivate reference curve R1s2 and activate R1s3 in the Plot Area 1 win-

dow, and run the model. The data for this run have 20% noise added. Edit the parameters that control deconvolution. The deconvolution smoothing factor, is set to a small value. Set it to a large value, such as 10000, and run the model again. How is the solution different? Try an intermediate value. Does this give a better compromise between the smoothness of the curve and the decreased peak height?

Return the deconvolution smoothing factor to a value of 10. Change to the fourth reference curve. Set the reference curve index to 4 in the Deconvolution window and activate reference curve R1s4 in the Plot Area 1 window. Run the model and observe the results when data with 40% noise are used. Can this fit be improved by increasing the value of the smoothing controller?

Summary

The results obtained when MMID4 (or any other model) is used as an analysis tool depend heavily on the quality of the reference data. It is essential to have data that are as close to error free as possible. This means that the experimental design must provide for adequate sampling, and care must be taken during all phases of tracer analysis to ensure that maximum accuracy is obtained. In addition to outflow curves, data acquisition should include measurement of flow, the distribution of flow, and, if at all possible, the input function.

3. MMID4 REFERENCE

3.1. Overview

3.1.1. *Model structure*

MMID4, represented diagrammatically by Fig. 1.1, models the transport and exchange of three tracers in an organ. It is comprised of discrete submodels that reflect the organ structure or the parts of the experimental apparatus that affect tracer concentrations and residues in the organ. These submodels represent nonexchanging vessels (such as tubes, arteries, arterioles, venules, and veins, modeled with vascular operators) and exchanging vessels (capillary-tissue units modeled by blood-tissue exchange, BTEX, operators).

For each tracer, there is one inlet tube, one artery, one vein, and one outlet tube. There may be a maximum of twenty flow pathways each consisting of an arteriole, a blood-tissue exchange unit, and a venule. Each pathway has its own flow rate and receives a specific fraction of the total mass of the tracers; additionally, each pathway may have its own permeability-surface area products (*PS*) and consumptions (*G*).

3.1.2. *MMID4, XSIM and the parameters*

MMID4 runs under XSIM. In this manual, when a description of a parameter is given, it will be given as in the following example:

Name	Description
Parameter name	Parameter usage

Input parameters (i.e., those for which the user must enter a value) are shown in normal type, while output parameters (i.e., those into which MMID4 writes values) are shown in *italics*. For example:

Name	Description
Fp	Mean plasma flow (ml min^{-1} g^{-1})
Cout_p	*Permeant tracer output concentration*

The first parameter is named “Fp”, and it contains the plasma flow. The user must enter the value of this parameter. The second parameter is named “Cout_p”. It contains the concentration of the permeant tracer at the outlet of the organ; the value of this parameter is set by MMID4 when a simulation is run.

The names shown are those that appear in the popup window after double clicking on a parameter button or field.

The XSIM convention for inputs and outputs is that input parameters are shown with a lighter field background, and the outputs are displayed on a shaded background, as shown in Fig. 3.1.

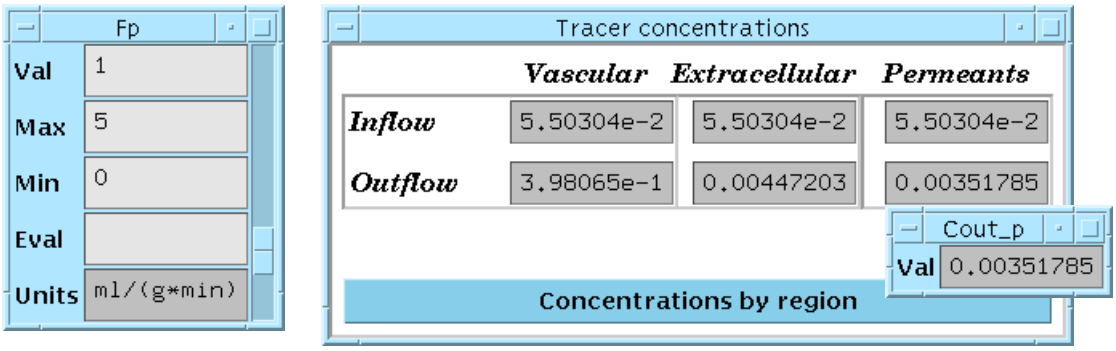


Figure 3.1. XSIM input parameters (left window) and output parameters (right windows).

3.1.3. *Using the MMID4 reference*

In addition to this overview, the MMID4 Reference Guide is divided into five parts. *Input Functions* (Section 3.2) deals with the several ways of getting the driving function, or functions, for the model. *Heterogeneity* (Section 3.3) deals with specifying heterogeneity of flow, *PS*, and consumption. *Vascular and BTEX Operators* (Section 3.4) deals with setting values for the transport and exchange parameters for the vessel operators. These sections are all concerned with specifying the inputs to MMID4. The final section, *Displaying Results* (Section 3.5), deals with the outputs from the model.

Each of these sections begins with a table that lists the model parameters that apply to the topic being discussed. This is followed by subsections that deal with the various aspects of the topic. The sections conclude with a listing and discussion of pertinent error messages and some tips on correcting error conditions.

3.2. Input Functions

3.2.1. *General*

As shown in Table 3.1, four options are available for selecting input functions. Input functions can be taken from the reference data curves stored in TAC (Time-Activity Curve) files; they can be generated by deconvolving a vascular output curve from the reference data. Additionally, a function generator can generate twelve different types of input functions. Regardless of the method used to select an input function, the data need not be at the same time points as those used for the simulation. For all input functions, linear interpolation is used to get the actual values used as inputs to the model.

Table 3.1: Options available for tracer input functions

1.	Use reference data curve
2.	Use deconvolution (Vascular tracer only)
3.	Use the function generator
4.	Use scaled vascular curve (Extracellular and permeant tracers)

3.2.2. *Using reference data curves*

Data files for XSIM, and thus for MMID4, are stored in a specialized file format known as TAC (Time-Activity Curves) and will be referred to as "reference data." The format of TAC files is described in the UNIX man page. (Enter "`man tac`" at the system prompt for details). A TAC file consists of one or more experimental runs. Each run may contain auxiliary data, physiological data, input data, and sample (or output) data. XSIM permits up to 10 input curves and 20 output curves to be loaded from the reference data file. XSIM will allow the user to see a summary of the TAC file structure (*Reference: TAC contents*), but this feature is currently not fully implemented.

To select a curve from the reference data to be used as the input function for a specified tracer, open the appropriate input window (e.g. *Parameters: input functions> Permeant input*), then press the *Select* button to choose Reference TAC. Click with the right mouse button in the TAC field, and select the appropriate input curve from the pop-up window. Entering a positive value in the Delay field will shift the curve to the right, and a negative value will shift it to the left.

Example 1

Fig. 3.2 shows the vascular tracer input taken from reference data curve #1, the extracellular input from curve #2, and the permeant input from curve #3. Input function scaling must be turned off, or else the extracellular and permeant curves will be slaved to the vascular input. See Section 3.2.5 for more information about input function scaling.

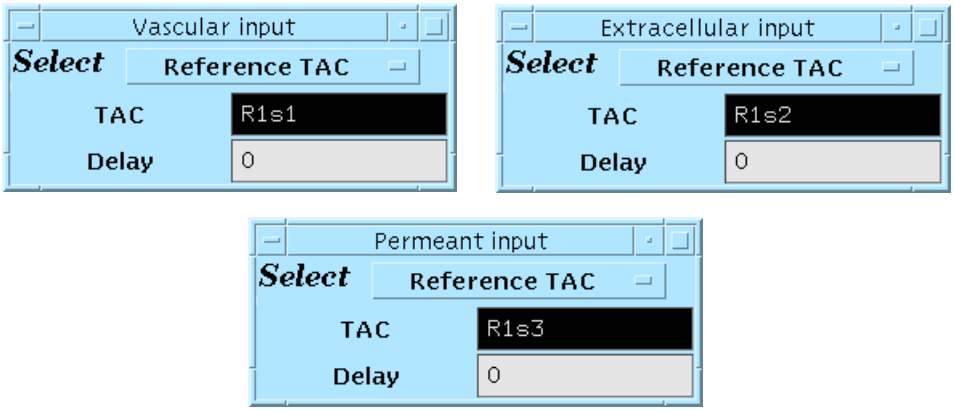


Figure 3.2. Reference data used as inputs for all three tracers.

Example 2

The vascular tracer input is taken from the reference data curve #1 and is scaled by 1.0. Because input function scaling is being used, the extracellular and permeant inputs are a 0.75 scaled version of the vascular input.

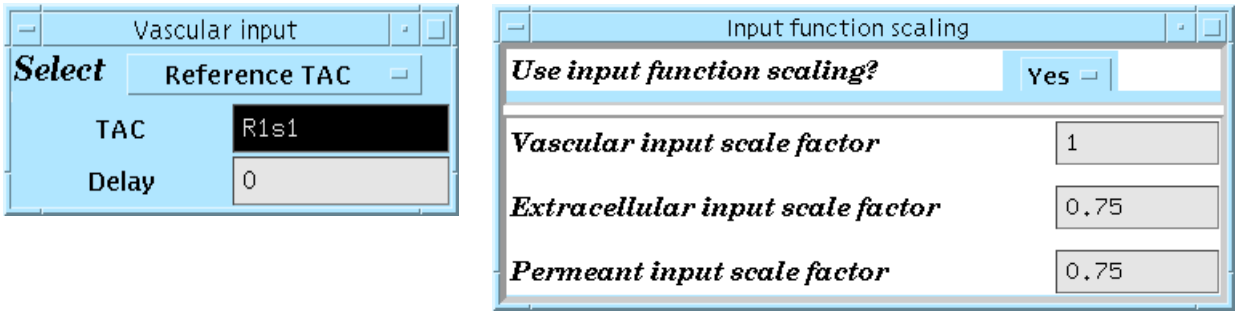


Figure 3.3. Extracellular and permeant inputs as a scaled function of the vascular input.

3.2.3. *Using deconvolution to generate a vascular tracer input curve*

Introduction

Deconvolution uses a vascular tracer output data curve and the transfer function of the vascular model (dependent upon the flow heterogeneity, large vessel volumes and dispersions, and vascular BTEX parameters) to calculate a vascular input curve. An example of deconvolution is depicted in Fig. 3.4.

The deconvolution process is significantly affected by noise in the measured data. The algorithm used for deconvolution attempts to minimize the inaccuracies caused by noise, and several smoothing parameters, discussed below, are under control of the user. Nevertheless, recording the vascular input at the time of the experiment will nearly always be preferable to using deconvolution. In some experiments, however, it may be impossible to record the signal, and deconvolution is required.

Deconvolution applies only to the vascular tracer input. The scaled results may, however, be used for the other tracers. This is appropriate when all the tracers are injected simultaneously and have the same waveform.

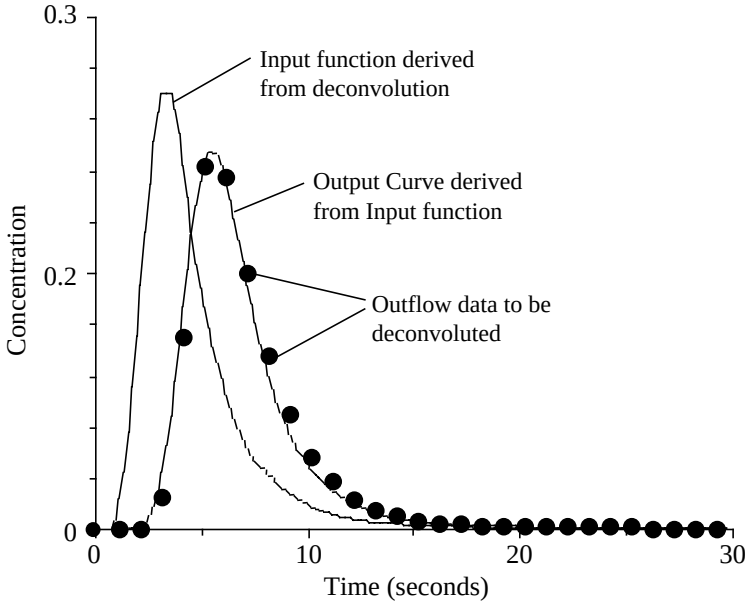


Figure 3.4. An example of deconvolution.

Selecting deconvolution

To calculate an input function by deconvolution, select *Parameters: Input functions> Deconvolution*, and set *Use deconvolution* to 'yes'.The deconvolution calculations can take considerable time; thus, when you next run the model, you may notice a significant delay prior to any results being plotted. To save time on subsequent runs, MMID4 stores and, as long as the vascular model parameters or the deconvolution parameters are not altered, reuses the last deconvolution results. Whenever the vascular model (time step, volume, flow, flow heterogeneity, diffusion coefficients, etc.), the deconvolution parameters, or the reference data file name are changed, the deconvolution is recalculated.

Selecting the data curve to be deconvoluted

The *Reference curve index* field specifies the data curve to be deconvolved. This index refers to the TAC file curve index, either an input curve index or a sample curve index, depending on the setting of the *Reference curve type* button. It is important to insure that the curve selected is actually a vascular tracer outflow curve or the results will be erroneous. (Usually the curve index is set to 1 for a vascular reference curve labeled as sample data, *e.g.*, an albumin concentration output curve.)

Deconvolution control parameters

Four parameters are available to the user to control the deconvolution process.

Deconvolution smoothing factor is an overall smoothing parameter for the deconvolution process, which controls the scaling of regularization matrices. This factor should be set to 1 for noise-free data, and in the range of 10 to 100 for noisy data. Permissible values are 10^{-5} to 10^{5}. The degree of smoothing increases as the smoothing factor value increases.

Optimize smoothing factor? If this button is set to 'yes,' then the deconvolution smoothing factor will be optimized. The optimization minimizes both the coefficient of variation between the output data and the deconvolved output and the excess variation of the estimated input function. If the optimizer

returns a smoothing factor of the order of 1000 or more, the data curve may not be suitable for deconvolution with this routine. See the manual page for more information (Type `man dcnopt` at the system prompt).

Data tail extension, controls smoothing of the tail of the derived vascular tracer input curve. *Time to start tail extension* specifies the time at which the power law or multi-exponential fit is applied.

Extracellular and Permeant scale factors are scalars for the two deconvolved curves.

Example

An example window configuration to deconvolve the reference data output curve #1 is given in Fig. 3.5.

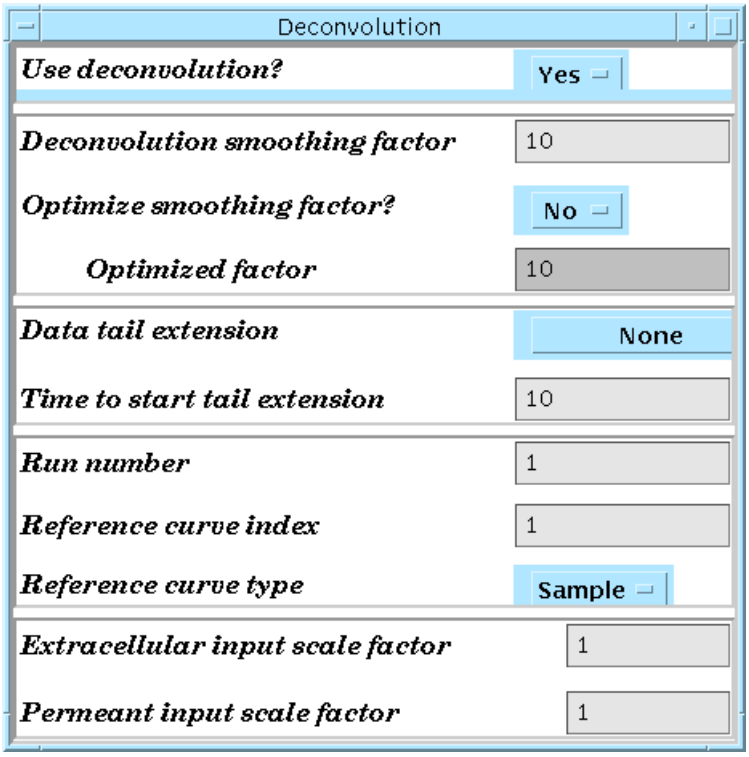


Figure 3.5. Configuration to obtain a deconvoluted input from the first reference data output curve.

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3.2.4. *Using the function generator to generate a tracer input curve*

Introduction

The function generator provides a variety of waveforms for creating input functions for all three tracers. The inputs for each of the three tracers may be independent, or they can be scaled to the vascular input by using input function scaling, described in the next section. Detailed information about the function generator subroutine, named cinput, can be found in the online UNIX manual page (`man cinput`).

Selecting a function generator

To select a function generator, open the desired input window and click on the *Select* button. In the following example, *Parameters: Input functions> Extracellular inputs* was used. Next, choose one of the functions by selecting from the Function button menu. In the example Fig. 3.6, a pulse input of 1 second duration has been chosen.

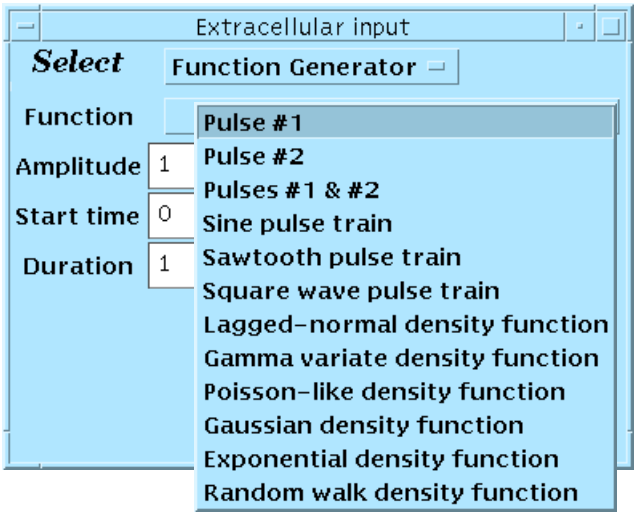


Figure 3.6. Function generator options with a pulse input function selected.

Example

In the example of Fig. 3.7, a lagged normal density curve (LNDC) has been specified for the vascular input. The LNDC has an area of 1.0, a mean of 5 seconds, a relative dispersion of 0.3, and a skewness of 1.2. Linearization of the upslope has been specified.

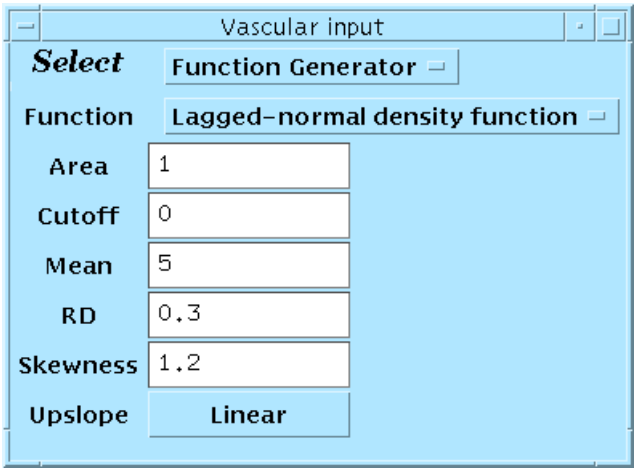


Figure 3.7. Lagged-normal density function with linear upslope.

3.2.5. *Scale inputs to the vascular curve*

If input function scaling is being used, then the input for all three tracer regions will be a scaled version of the vascular input function. Otherwise, the

inputs for each region are independently set. For each tracer input, the *scale factor* provides a scaling factor applied to the values generated by the function selection. It must have a nonnegative value.

Note that if the vascular scaling factor is changed, only the scaling of the vascular input function changes, while the scaling of the other two inputs is not affected. The extracellular and permeant tracer inputs are affected only by their respective scaling factors.

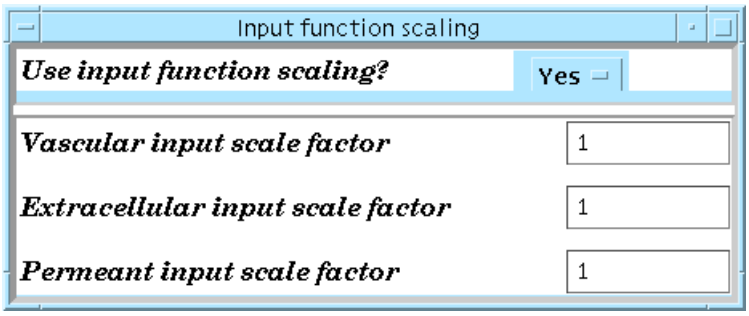


Figure 3.8. Input function scaling.

3.2.6. *Input function errors and messages*

The input function errors and messages are largely self-explanatory.

3.3. Heterogeneity

3.3.1. *General*

Organs are known to be heterogeneous in many of their physical properties including regional blood flow. In MMID4, flow heterogeneity is modeled by a set of parallel flow paths as shown in Fig. 1.1. Each of the up to 20 pathways has a different blood flow. The procedure used to specify the blood flows is discussed in the next section.

In addition to heterogeneity of regional blood flow, MMID4 also permits heterogeneity in membrane conductances and consumptions (Section 3.3.3). If heterogeneous conductances and/or consumptions are used, the values for each path are proportional to the flow for that path. Thus, these heterogeneities can only be used in conjunction with heterogeneity of flow.

The parameters and windows related to heterogeneity are shown in Fig. 3.9. The number of paths, found in the MMID4 model layout window, is the primary controller for heterogeneity. As the name implies, it sets the number of flow pathways used by the model. If it is set to 1, no heterogeneity is used regardless of the values of the other heterogeneity parameters.

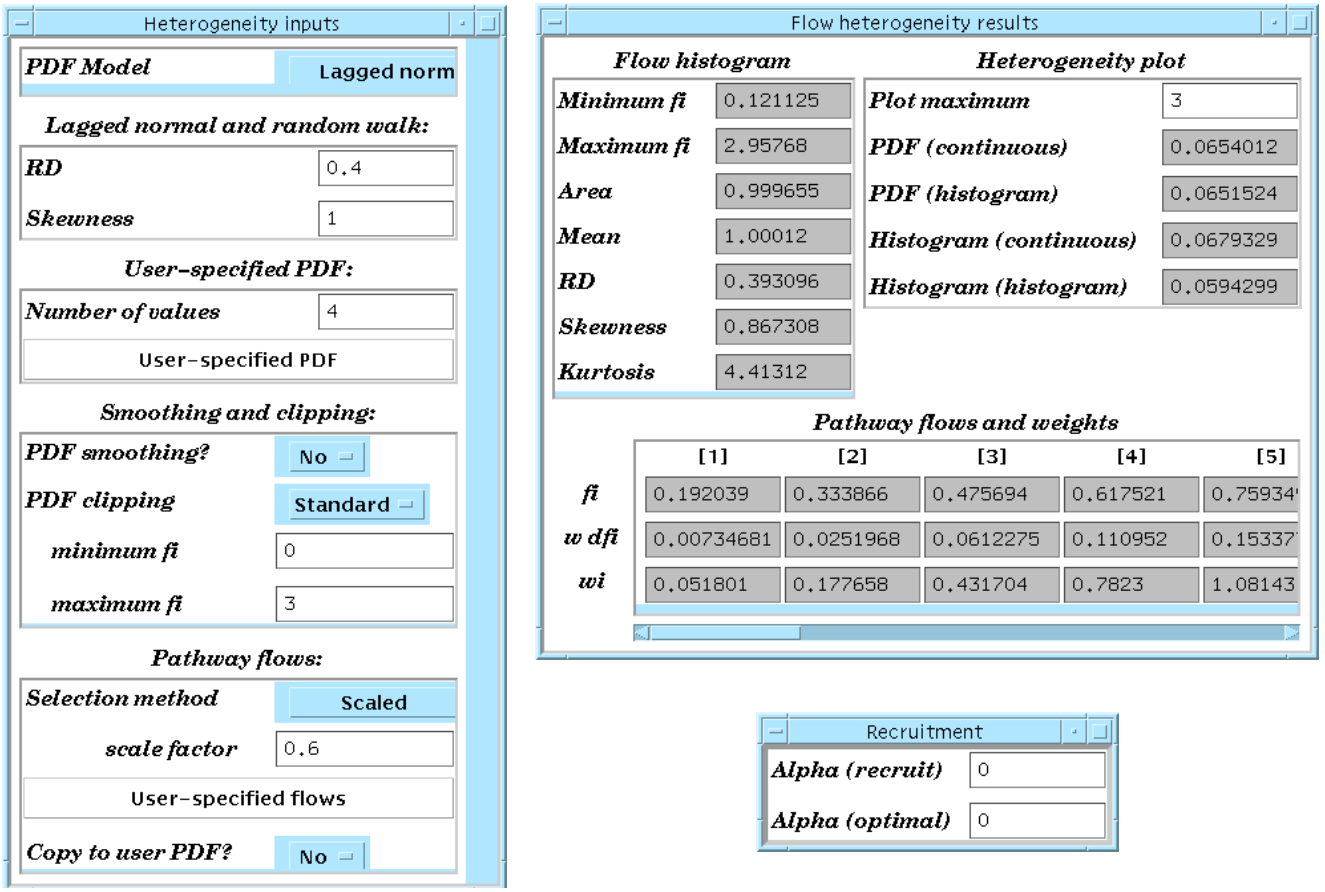


Figure 3.9. Heterogeneity input, output and recruitment windows.

3.3.2. *Flow Heterogeneity*

Introduction

Blood flow heterogeneity affects the shape of outflow and residue curves measured in indicator dilution experiments. Comparing the outflow curve from a multipath (heterogeneous flow) model to that from a single path (homogeneous flow) model, differences include

1. shortened appearance time (caused by the presence of paths with flows greater than the mean),
2. decreased peak height and increased dispersion (caused by distributing the total flow among paths with different flows), and
3. an elevated tail of the curve (caused by paths with flows lower than the mean).

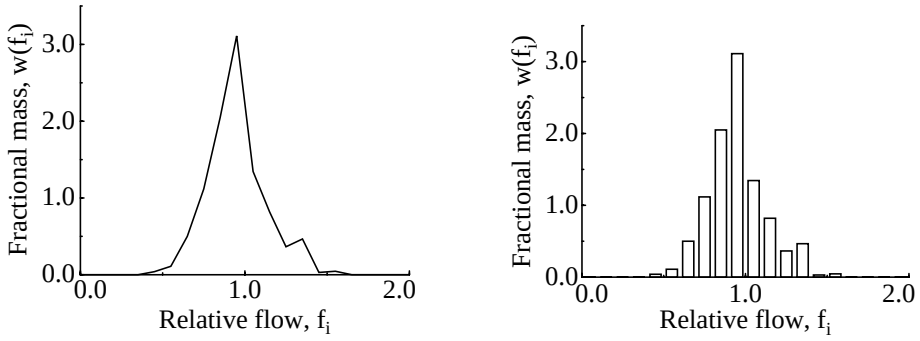


Figure 3.10. Probability density function (PDF) of local regional flows. *Top:* Continuous representation. *Bottom:* Histogram representation.

Regional blood flow in an organ can be described by a probability density function (PDF) of relative flows. (The relative flow is the regional flow divided by the mean flow to the organ.) A sample regional flow PDF, is shown in Fig. 3.10. The relative flow, f_i, is the regional flow divided by the mean flow for the whole organ, and the fractional mass, $w(f_i)$, is

$$w(f_i) = \frac{\sum m_j}{W \cdot \Delta f_i} \tag{3.1}$$

where Σm_j is the total weight of all the organ regions with relative flows in the i^{th} flow class, W is the total weight of the organ, and Δf_i is the width of the i^{th} class. The PDF has a mean of 1.0, and an area of 1.0. As shown in Fig. 3.10, the PDF may be represented as a histogram or as a continuous function. The former has the advantage of showing the widths of the flow classes, while the latter allows more than one PDF to be displayed on the same axes. In this example, all the flow classes have equal widths, but this need not be the case.

Terminology used for flow heterogeneity

In MMID4, each flow class (maximum of 20) is modeled by a separate flow path. The relative flow for the path, f_i, is referred to by $f(N)$, and the fractional mass, $w(f_i)$, is referred to as $w(N)$, where N is the path number.

Note that the area of the PDF is unity. Thus $\sum(w_i \cdot \Delta f_i) = 1.0$. Since Δf_i need not be constant for all pathways, it may be difficult to interpret a set of w_i's by visual inspection. To overcome this difficulty, the weighting factors for the paths in MMID4 are specified in terms of the $w_i \Delta f_i$ for that path, referred to as $wd(N)$. Thus $wd(N)$ is the fraction of the organ represented by the flow for the N^{th} pathway. For a properly normalized PDF, the sum of the $wd(N)$'s is 1.0.

As described below, the user may specify the PDF relative flows and weighting factors in the parameter array. These are referred to as "*Fin*" and "*WDin*" respectively and are accessed by clicking on the User specified PDF button. The user may also specify a set of relative flows to be used for the paths; these are referred to as "*Fout*" and their window is opened by pressing the User-specified flows button. The relative flows and weights for the paths that are actually used by the model are referred to simply as *fi* and *wdfi* and are located in the Flow heterogeneity results window. The fractional mass values, *wi*, for the paths are also calculated and are located in the same results window.

Calculation of flow heterogeneity parameters

The procedure used in MMID4 for calculating the heterogeneity parameters from the user specified values is diagrammed in Fig. 3.11. The goals are to allow several choices for selecting a PDF that represents the distribution of flows in the tissue being modeled, allow flexibility in selecting the flows used for the pathways, and select weights for these flows that, in so far as possible, faithfully represent the selected PDF. In Phase 1, a PDF that characterizes the organ being modeled is specified. Relative flows that best represent the data being analyzed and weights for each path are selected/calculated in Phase 2.

Three choices are available in Phase 1a:

1. the PDF for a particular experiment can be loaded from the reference data,
2. a PDF can be taken from the parameter array, or
3. a mathematical distribution can be used.

Two mathematical distributions are available: a lagged normal density curve (LNDC) or a random walk (RANWOK). The LNDC is used for distributions with moderate skewness and RANWOK is used when the skewness exceeds three times the relative dispersion. In Phase 1b, the high and low ends of the PDF are clipped to eliminate paths with very small probabilities (default $wd_i < 0.1\%$). The user may specify additional clipping.

In Phase 2a, the relative flows that will be used by the model, f_i, are selected. These flows can be specified by the user or generated by a mathematical algorithm that selects flows equally spaced in either the flow or the transit time domains, or at some point intermediate between those limits. If the flows are specified by the user, they may be scaled and shifted to bring them into the range of flows spanned by the PDF. In Phase 2b, the PDF is interpolated to obtain the weighting factors for the paths, wd_i. Finally f_i and wd_i are normal-

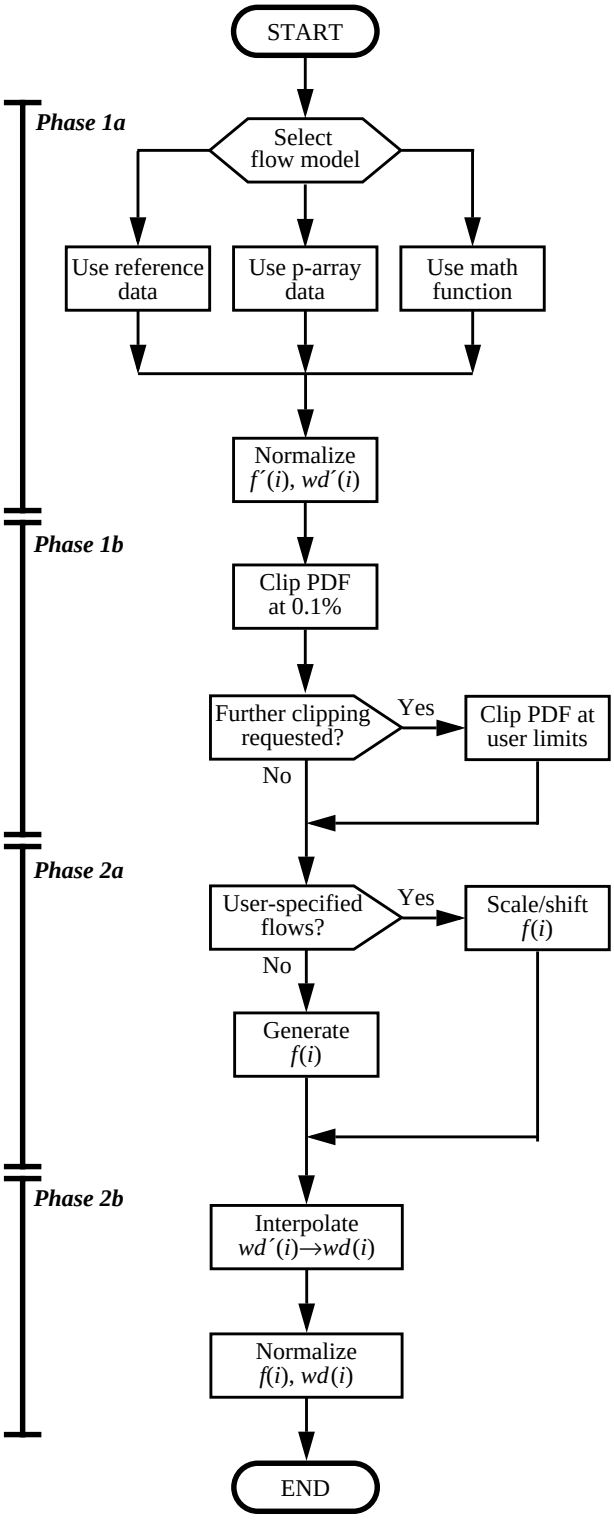


Figure 3.11. Procedure used in MMID4 for calculating relative flows, $f(i)$, and weighting factors, $wd(i)$, from user specified parameters.

ized to ensure that mass is conserved in the model. Not shown on the diagram is the calculation of the w_i's that correspond to the wd_i's.

While considerable emphasis has been placed on faithfully representing the specified PDF by the relative flows and weights used for the paths, the procedure is not exact. Thus, it is ***strongly*** recommended that the user plot the PDF's as described below and visually compare the results. (See *Plotting the regional flow PDF's*.)

Specifying the heterogeneity model (Phase 1a)

Selection of the heterogeneity model is controlled by the *PDF Model* selection button in the Heterogeneity inputs window.

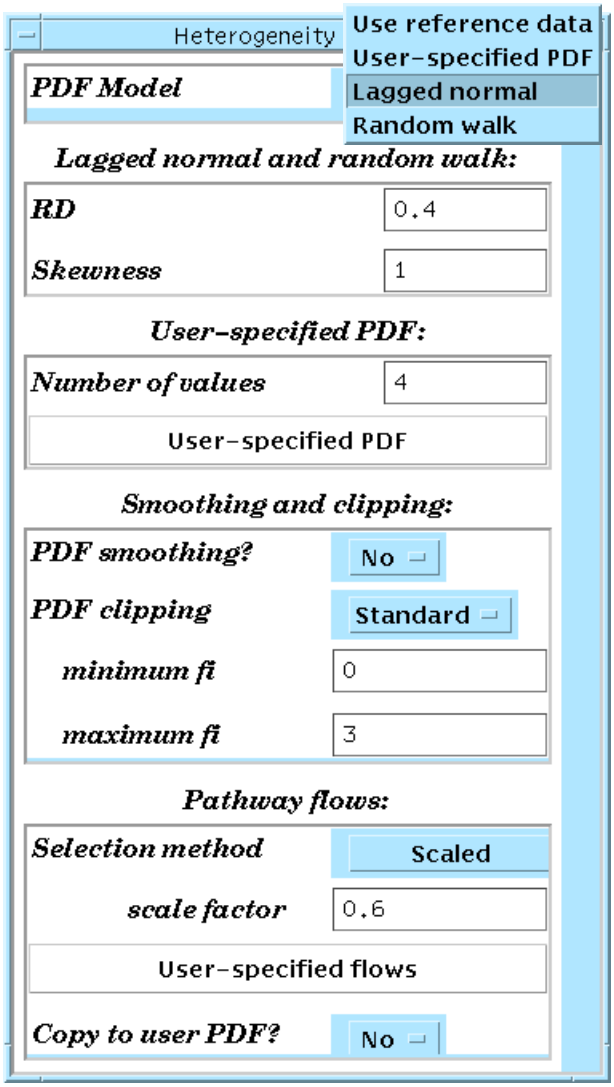


Figure 3.12. Heterogeneity inputs window.

Using a PDF from the Reference data

If Use reference data is selected, the PDF will be read from the reference data. The data are loaded into the area reserved for a user-specified PDF discussed

below. All restrictions and considerations related to a user-specified PDF apply. Note that any data in the user-specified PDF locations are overwritten.

Using a user-specified PDF

User-specified PDF: In keeping with established practice at the NSR labs for standardizing experimentally determined flow PDF's, a maximum of 30 values are permitted. The number of values actually used is specified in the *Number of values* field. The f_i's and wd_i's are shown in the User-specified PDF pop-up window, *Fin* and *WDin*, respectively.

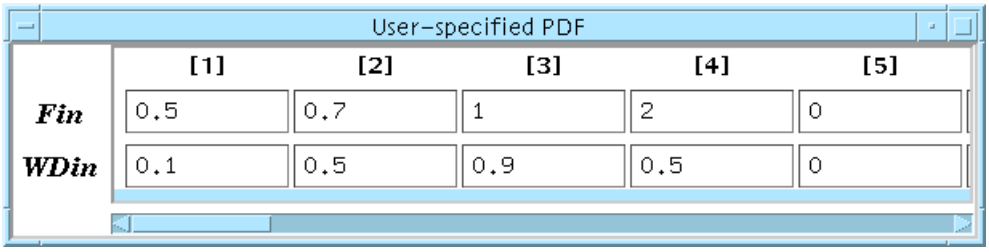


Figure 3.13. User-specified PDF window.

These data can be directly edited by the user to change the shape of the PDF. The values need not be normalized to an area and mean of 1.0 as MMID4 will normalize the PDF before using it in any further calculations. Note that the values in the parameter array will be overwritten if a reference data PDF is used or if the *Copy to user PDF?* switch, discussed below, is on.

Smoothing the PDF

A reference data or user-specified PDF may be smoothed prior to its usage in further calculations. Smoothing is turned on by setting the *PDF smoothing?* switch to Yes. If smoothing is on, a running three point smoother with weights [0.25, 0.50, 0.25] is used.

Using a mathematically generated PDF

If no PDF is available from the reference data and usage of a user-specified PDF is not desired, two mathematical functions are available, a lagged normal density function and a random walk density function. The shape of the PDF is controlled by these parameters:

Name	Values	Usage
RD	0.0–1.0	Relative dispersion
Skewness	See note below	Skewness

Permissible values of the skewness depend on the function selected. For LNDC, skewness must be in the range 0.0–1.9. For Random walk, skewness must be greater than 3.0 * *RD*.

Since negative values of relative flow are not permitted, using large values of *RD*, especially in conjunction with a small value of skewness, will result in the PDF being significantly clipped on the low end. Examples of PDF's generated with LAGNDC and RANWOK are shown in Fig. 3.14.

Error conditions

If an error is detected in the parameters that control the heterogeneity model, a warning message is issued and a lagged normal distribution is used.

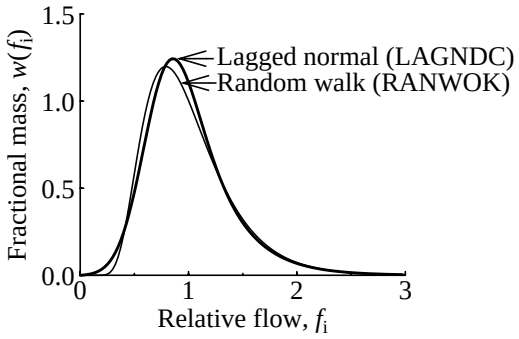


Figure 3.14. Mathematically generated PDF's. *RD* = 0.4; skewness = 1.2.

Clipping the PDF tails (Phase 1b)

To avoid useless calculations, flow paths with low weights are removed by clipping the tails of the PDF. By default, standard clipping is done at the points at which wd_i is less than 0.001. The user may specify custom clipping that is controlled by these parameters:

Name	Values	Usage
PDF clipping	Standard	Clip PDF at default limits
	Custom	Clip PDF at limits specified by fmin and fmax
Minimum *fi*	< 0.5	Minimum f_i for PDF
Maximum *fi*	> 1.5	Maximum f_i for PDF

The clipping limits, *minimum fi* and *maximum fi*, must be less than 0.5 and greater than 1.5 respectively. If either is outside its range, it is reset to the limit and used in the subsequent calculations. The altered value is stored in the parameter array and a warning message is printed.

Additional clipping should be used with caution. Since the results will be renormalized in Phase 2b, excessive clipping can result in a final distribution that has a significantly altered shape and statistics than that originally specified.

Specifying the relative flows for the paths (Phase 2a)

The specifications of the f_i's in Phase 1 are used to determine the shape of the PDF. In Phase 2a, the relative flows that are actually used for the paths are selected, as determined by the pathway flows *Selection method*.

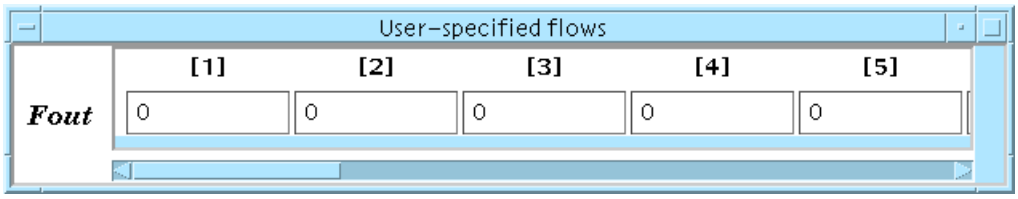


Figure 3.15. User-specified flows window.

If user-specified flows are selected, the values to be used are entered in the User-specified flows pop-up window. Up to 20 values may be specified, but the

number actually used is determined by the number of paths selected in the MMID4 model layout window. The values must be greater than 0.0 and in ascending order. If an error is detected, a warning message is printed and weighted spaced flows are used as described below. If some of the user specified flows fall outside the clipping limits specified above, then the set of user-specified flows are scaled and/or shifted so that they fall within the clipping limits. This preserves the relative spacing of the flows over the clipped flow range.

If mathematically generated f_i's are selected, the user can choose from four different methods of spacing the f_i's:

1. equally spaced in the flow domain, Equal F Spacing (Fig. 3.16a, b),
2. equally spaced in the transit time domain, Equal t spacing (Fig. 3.16b, c),
3. intermediate between (1) and (2), Scaled (Fig. 3.16d, e), or
4. weighted flows, Weighted (Fig. 3.16f, g).

Equal flow domain spacing results in many paths with short transit times and, thus, few paths with long transit times. This may lead to deformation of the tails of outflow curves due to undersampling of long transit times. Equal flow domain spacing almost always preserves the statistics of the PDF selected. Equal transit time spacing inadequately preserves the PDF statistics, but may give a better representation of the tails of the curves. Using a *scale factor* between 0 and 1 gives f_i's that are linearly interpolated between the equal flow and transit time spacing. Fig. 3.16d, e show the results when *scale factor* is set to 0.6. Selecting the weighted method selects flows that are weighted to be equally spaced in transit time at low flows and equally spaced in flow at high flows as seen in Fig. 3.16f, g. This choice is recommended as it preserves the statistics of the flow PDF and gives the more flows at the ends of the PDF that effect the upslope and tail of the curves.

Generating the weights for the flow paths (Phase 2b)

The weights for the pathways are obtained by interpolating the PDF generated in Phase 1 at the values of relative flow generated in Phase 2a. After the interpolation, the results are renormalized to insure conservation.

Flow heterogeneity results

The normalized PDF resulting from Phase 2b is stored in the Flow heterogeneity results window, Fig. 3.17. Relative flows, f_i, path weights, w_i, and fractional masses, $wd(f_i)$, are displayed. The flow limits and the statistics of the PDF are shown in the *Flow histogram* section.

Copying the heterogeneity results

The relative flows and weights used for paths, *fi* and *wdfi* in the results window, can be copied to the User-specified PDF window, *Fin* and *WDin*. This action is controlled by the *Copy to user PDF?* switch in the Heterogeneity inputs window. When it is set to Yes, the heterogeneity results are copied to the user-specified PDF section. No action is taken when the switch is set to No.

If the results are copied, the number of user-specified values is set to the current number of paths. These data can then be edited and, if the pathway flows selection method is set to equal F spacing, used as the input PDF for subsequent model runs.

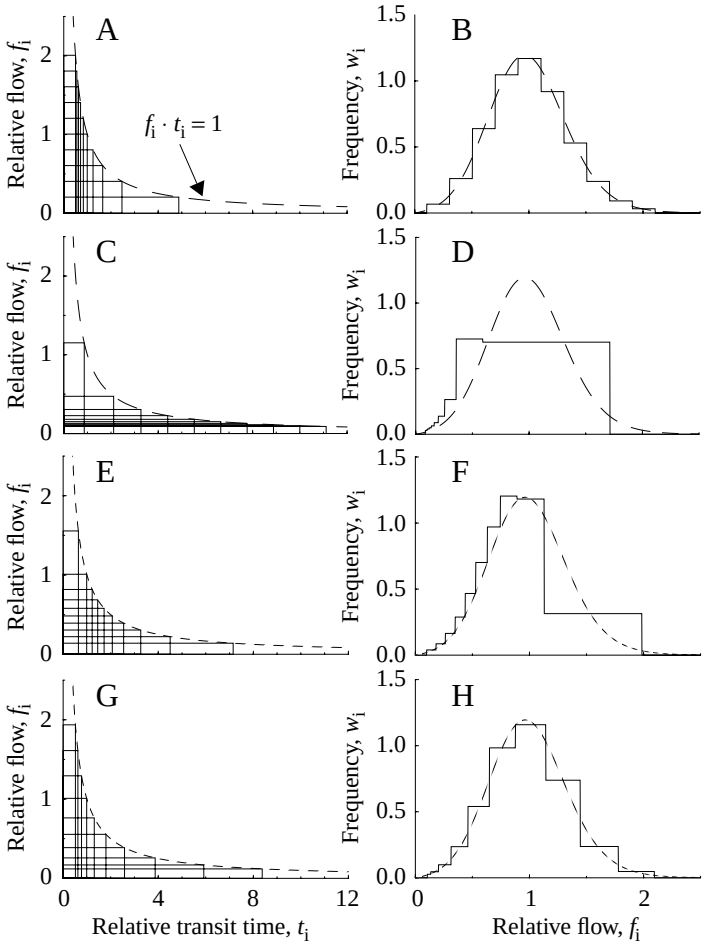


Figure 3.16. Methods of selecting relative flows for a flow histogram. *Left panels:* relative flows and transit times of pathways. Each relative flow, f_i, corresponds to a relative transit time, $\bar{t}_i = 1/f_i$. *Right panels:* The flow histograms, solid lines, result from selecting flows from the corresponding panels on the left. Four methods of selecting flows are shown: (1) equal flow spacing (a, b), (2) equal transit time spacing (c, d), (3) intermediate flows with a scaling factor of 0.6 (e, f), and (4) weighted flows (g, h). The flow PDF (LNDC with RD of 0.35 and skewness of 0.30) is shown by the dashed line.

When the copy is complete, the copy switch is turned off. This avoids continuous copying and renormalization of the PDF that can result in undesirable shifts in the relative flows.

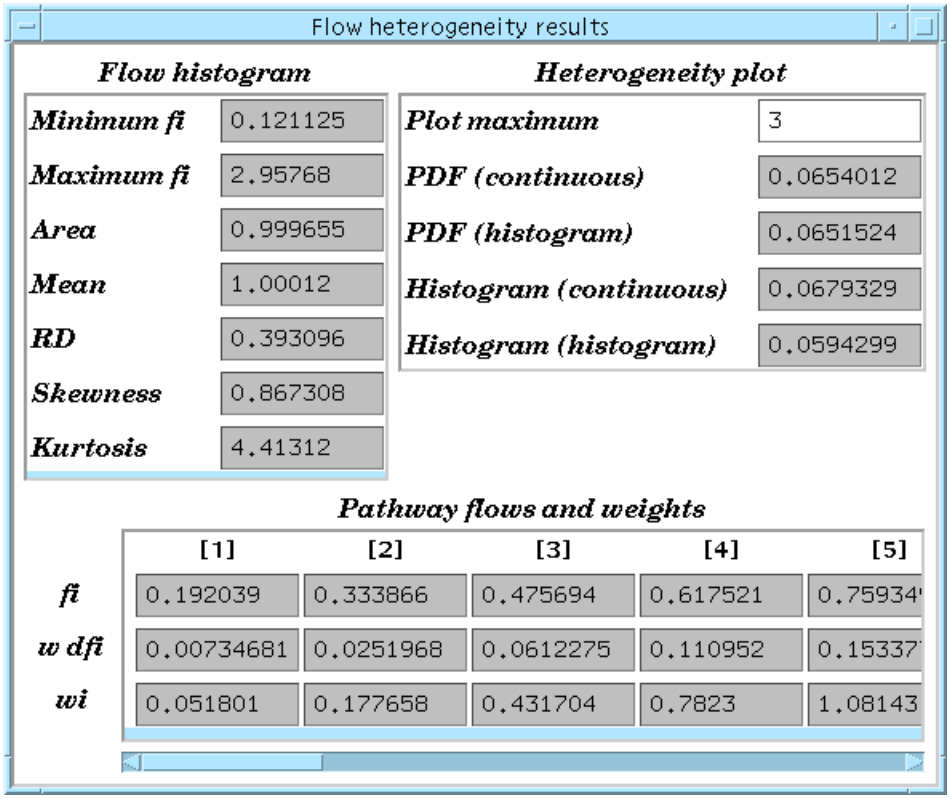


Figure 3.17. Flow heterogeneity results window.

Plotting the regional flow PDF's

The input PDF and the PDF used by the pathways is available for plotting. Relevant parameters are:

Name	Value	Usage
Plot maximum	...	Maximum flow value for x-axis. (Default = 3.0)
PDF_hist	...	Input PDF (histogram representation)
PDF_cont	...	Input PDF (continuous representation)
HST_hist	...	Model PDF (histogram representation)
HST_cont	...	Model PDF (continuous representation)

Parameters *PDF_hist, PDF_cont, HST_hist* and *HST_cont* are updated at each step of the simulation and can be plotted in the usual manner for XSIM outputs by entering the parameter name in a plot area Y parameter field. An example of plotting HST_hist and PDF_cont is shown in Fig. 3.18. It is recommended that the continuous version of the input PDF and the histogram version of the model PDF be plotted so that they can be easily distinguished.

The x-axis scaling is controlled by *Plot maximum*. The x-axis minimum is always 0.0. If *Plot maximum* is set to 0 or less, the default value of 3 is used for the x-axis maximum. (Note: The x-axis labeling is still controlled by the scaling of the independent variable.)

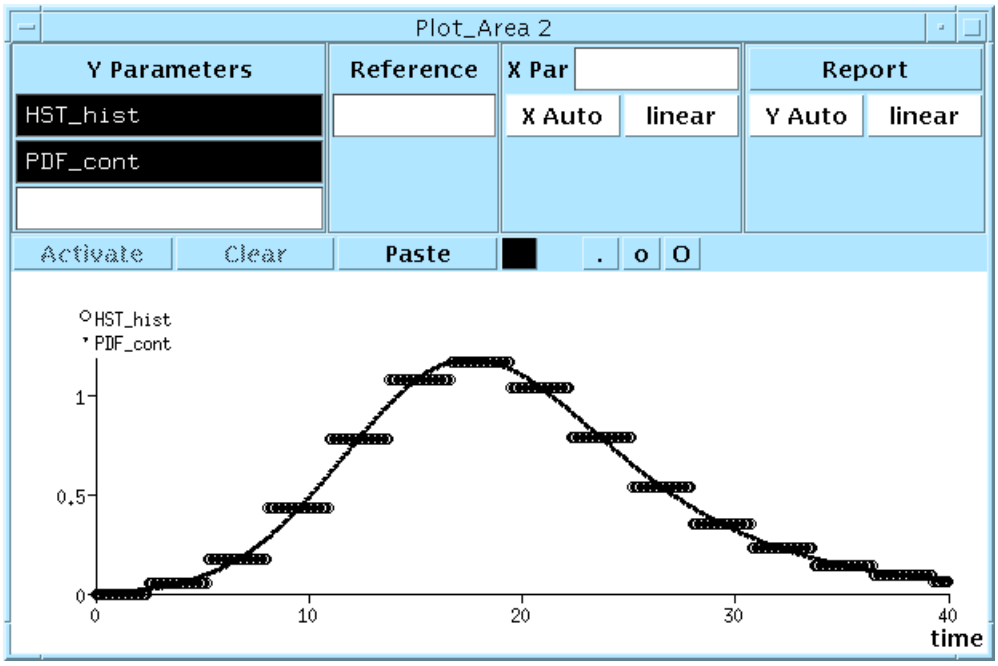


Figure 3.18. Plotting input and model PDFs.

3.3.3. *Heterogeneous membrane conductances and consumption*

Introduction

Recent studies have indicated, though not conclusively proven, that regional membrane conductances, permeability-surface area (*PS*) products, are related to regional flow. One mechanism for this is increased surface area as additional capillaries are recruited. Similarly, it may be expected that there is a relationship between the regional consumption, or utilization, of a substrate and its delivery to the region (i.e., some optimal balance between flow and consumption).

These effects are modeled in MMID4 by including mathematical relationships between flow and *PS* or *G* in each path. The function used is

$$P_i = P_{mean} \cdot (1 - \alpha + \alpha \cdot f_i) \qquad i = 1, \ldots, N_{path}, \tag{3.2}$$

where P is either *PS* or *G*, P_{mean} is the average value of the parameter specified in the parameter array, α is α_{rct} or α_{opt}, f_i is the relative flow for the pathway, and P_i is the actual value of the parameter used for that path. Note that $\alpha = 0$ makes the parameter constant for all path and $\alpha = 1$ makes the parameter directly proportional to flow.

These relationships are controlled by the parameters:

Name	Value	Usage
Alpha (recruit)	0.0 - 1.0	Proportionality constant for recruitment, α_{rct}
Alpha (optimal)	0.0–1.0	Proportionality constant for optimality, α_{opt}

Recruitment heterogeneity

Setting α_{rct} produces heterogeneity in PS_g for all tracers and in the endothelial cell permeabilities, PS_{ecl} and PS_{eca}, for the permeant tracer.

Optimality heterogeneity

Setting α_{opt} only affects the parameters for the permeant tracer. It produces heterogeneity in PS_{ecl}, PS_{eca}, PS_{pc1}, PS_{pc2}, G_{ec}, and G_{pc}. Note that α_{rct} and α_{opt} both affect PS_{ecl} and PS_{eca} for the permeant tracer. If α_{opt} is nonzero, it overrides the control specified by α_{rct}.

The Recruitment window is accessed from the *Parameters* pull-down menu.

Figure 3.19. Recruitment window.

3.3.4. *Heterogeneity errors and messages*

[TO BE DEVELOPED]

3.4. Vascular and BTEX Operators

3.4.1. *Introduction*

The configuration of the circulatory operators in MMID4 is shown in Fig. 1.1. Of the operators shown, all nonexchanging operators, input tubing, artery, arterioles, venules, vein, and output tubing, are modeled with vascular operators. The exchange units are modeled with BTEX operators. Parameters related to the circulatory operators are those shown in Table 3.2.

Table 3.2: Circulatory operator parameters

Name	Description	
Fp	Plasma flow, ml min^{-1} g^{-1}	
Vp	Plasma volume, ml g^{-1}	
N paths	Number of pathways (See Section 3.3)	
N segments	Number of axial segments	
cap length	Capillary length, cm	
Vtube_i	Input tubing:	Volume, ml g^{-1}
RDtube_i		Relative dispersion
Vart	Artery:	Volume, ml g^{-1}
RDart		Relative dispersion
Vartl	Arterioles:	Volume, ml g^{-1}
RDartl		Relative dispersion
Vvenl	Venules:	Volume, ml g^{-1}
RDvenl		Relative dispersion
Vven	Vein:	Volume, ml g^{-1}
RDven		Relative dispersion
Vtube_o	Output tubing:	Volume, ml g^{-1}
RDtube_o		Relative dispersion
v_Dp	Vascular tracer:	Axial diffusion constant in plasma, cm^2 sec^{-1}
v_PSg		PS for capillary-ISF exchange, ml min^{-1} g^{-1}
v_Visfp		ISF virtual volume, ml g^{-1}
v_Czero		Initial concentration, arbitrary units
e_Dp	Extracellular tracer:	Axial diffusion constant in plasma, cm^2 sec^{-1}
e_PSg		PS for capillary-ISF exchange, ml min^{-1} g^{-1}
e_Visfp		ISF virtual volume, ml g^{-1}
e_fVisf2		Fraction of ISF volume in ISF2

Table 3.2: Circulatory operator parameters

Name	Description	
e_Disf		Axial diffusion constant in ISF, $cm^2\ sec^{-1}$
e_PSisf		PS for ISF1-ISF2 exchange, ml $min^{-1}\ g^{-1}$
e_Czero		Initial concentration, arbitrary units
p_Gp	Permeant tracer:	Consumption in plasma, ml $min^{-1}\ g^{-1}$
p_Dp		Axial diffusion constant in plasma, $cm^2\ sec^{-1}$
p_PSecl		PS for capillary-endothelial cell exchange, ml $min^{-1}\ g^{-1}$
p_PSeca		PS for endothelial cell-ISF exchange, ml $min^{-1}\ g^{-1}$
p_Vecp		Endothelial cell virtual volume, ml g^{-1}
p_Gec		Consumption in endothelial cell, ml $min^{-1}\ g^{-1}$
p_Dec		Axial diffusion constant in endothelial cell, $cm^2\ sec^{-1}$
p_PSg		PS for capillary-ISF exchange, ml $min^{-1}\ g^{-1}$
p_Visfp		ISF virtual volume, ml g^{-1}
p_Gisf		Consumption in ISF, ml $min^{-1}\ g^{-1}$
p_Disf		Axial diffusion constant in ISF, $cm^2\ sec^{-1}$
p_PSpc -> isf1		PS for parenchymal cell-ISF1 exchange, ml $min^{-1}\ g^{-1}$
p_PSisf		PS for ISF1-ISF2 exchange, ml $min^{-1}\ g^{-1}$
p_fVisf2		Fraction of ISF volume in ISF2
p_PSpc -> isf2		PS for parenchymal cell-ISF2 exchange, ml $min^{-1}\ g^{-1}$
p_Vpcp		Parenchymal cell virtual volume, ml g^{-1}
p_Gpc		Consumption in parenchymal cell, ml $min^{-1}\ g^{-1}$
p_Dpc		Axial diffusion constant in parenchymal cell, $cm^2\ sec^{-1}$
p_Czero		Initial concentration, arbitrary units

3.4.2. *Plasma flow through vascular and BTEX operators*

The total plasma flow, ml $min^{-1}\ g^{-1}$, is set in the MMID4 model layout window:

Name	Values	Usage
Fp	>0	Plasma flow, ml $min^{-1}\ g^{-1}$

The total plasma flow is delivered to the input tubing, artery, vein, and output tubing. The fraction of this flow that is delivered to each pathway is controlled by the number of paths and the flow heterogeneity. All the flow delivered to a path goes to the arteriole, BTEX unit, and venule.

3.4.3. *Vascular operators*

Introduction

All nonexchanging vessels are modeled with vascular operators that provide delay and dispersion. Each operator is characterized by two parameters: volume, *V*, and relative dispersion, *RD*. Note that no consumption of tracer is permitted in these operators and that the parameters set for the vascular operators apply to the operators for all tracers.

The maximum value permitted for relative dispersion is 0.48, and the minimum value is 0. In the latter case, the operator acts as a pure delay line. If the specified value is outside the permitted range, it is reset to the nearest limit and a warning message is written.

The volume should be the physical volume of that component of the circulation in ml g^{-1}. If the volume of an operator is set to 0, it is effectively removed from the system, and its residue function will always have a value of 0 (See Section 3.5.4).

Detailed information about the vascular operator is contained in King, et al. (1993); information about the computer implementation is available on-line (`man vasop`).

Arterial operators

The vascular operators on the arterial side are the inlet tubing, artery, and arterioles.

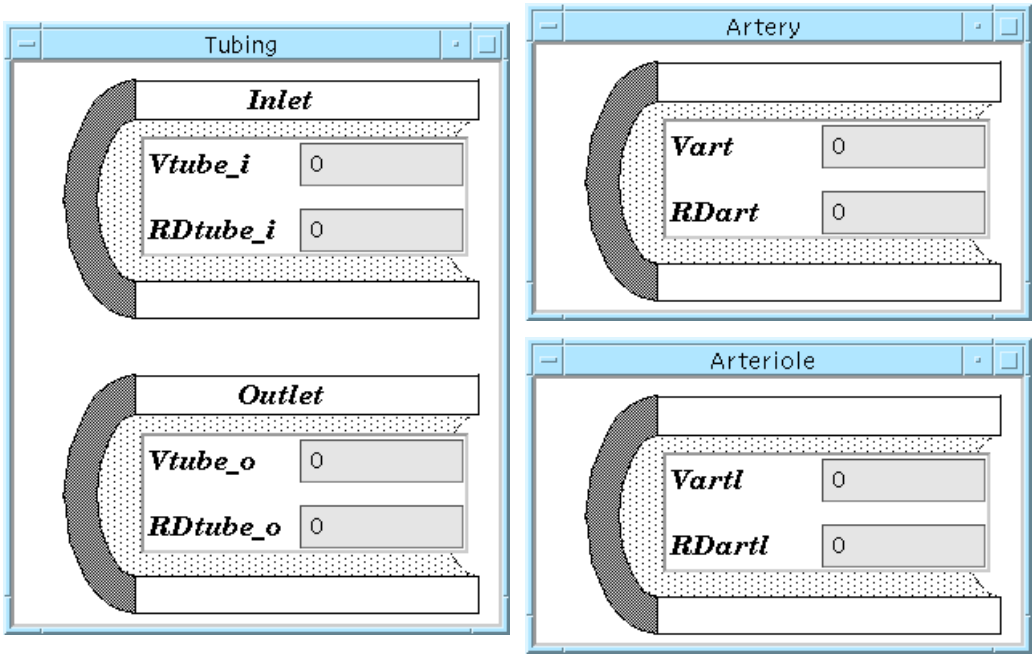


Figure 3.20. The tubing, artery and arteriole vascular operator windows.

For each tracer, there is a single inlet tube and artery. Separate operators are provided for these two components for use when the tissue content of the tracer

(residue) is being observed. The inlet tube can provide delay and dispersion of the tracer injection before it reaches the tissue that is included in the residue signal. (See Section 3.5.4.)

There is one arteriole for each of the flow paths. The volume specified for this component is divided equally amongst the paths.

Venous operators

The vascular operators on the venous side are the venules, vein, (Fig. 3.21) and outlet tubing (Fig. 3.20).

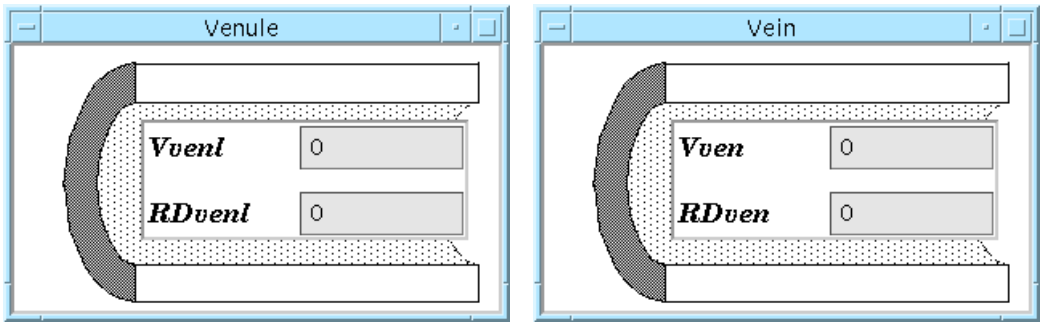


Figure 3.21. The venule and vein vascular operator windows.

As with arterioles, there is one venule per path, and the volume is distributed equally. The comments made above regarding the inlet tubing and artery also apply to the vein and outlet tubing.

3.4.4. *BTEX operators*

Introduction

The capillary-tissue units of each path are modeled with BTEX operators. A different type of BTEX operator is used for each tracer depending on the number of regions required to model the behavior of that tracer. Detailed information about the BTEX operators is contained in Bassingthwaighte, Chan and Wang (1992); information about the computer implementation of a typical three region model is available on-line (`man btex30`).

The generic parameters of a BTEX model are shown in the table below. The names used in MMID4 are prefixed with the tracer label (*v:* vascular tracer, *e:* extracellular tracer, *p:* permeant tracer, *no prefix:* all tracers) and postfixed by the region label (*p* plasma, *ec* endothelial cell, *isf* interstitial fluid, and *pc* parenchymal cell). The full XSIM variable name is given in parenthesis if it differs from the name that is initially visible in most windows.

Name	Usage
V	Volume of the region. Note that for all extravascular regions (*ec*, *isf*, and *pc*) the volume is a volume of distribution rather than a physical volume.
D	Axial diffusion constant in the region.
PS	Permeability-surface area product for radial diffusion into adjacent regions.
G	Consumption of tracer (gulosity) in the region.

Name	Usage
*C*zero	Initial concentration of tracer in all regions.

Common parameters

Some BTEX parameters are common to all the tracers. These common parameters, with the valid range or typical values, are

Name	Value	Usage
Fp	1.0	Plasma flow, ml min^{-1} g^{-1}
Vp	0.03	Plasma volume, ml g^{-1}
Path (Npath_int)	1–20	Number of flow paths
Nseg (Nseg_int)	1–60	Number of axial segments
capillary length (clngth)	0.1	Capillary length, mm

Vascular tracer

The vascular tracer is modeled with a two region BTEX model (Fig. 3.22, top). While the vascular tracer is nominally constrained to the vascular region, the second region is used to account for any binding of the tracer to the endothelial cell. An example of this effect is binding of albumin to an albumin-fatty acid receptor on the endothelial cell membrane. If appropriate, this second region can also be used to account for slow leakage of the vascular tracer into the ISF over long times.

Typical values of the vascular tracer parameters are:

Name	Value
v_Dp	0.0
v_PSg	0.0
V'isf (v_Visfp)	0.0
v_Czero	0.0

Extracellular tracer

The extracellular tracer also has access to two regions, plasma and ISF (Fig. 3.22, middle). For this tracer, however, the ISF is partitioned into two parts. This partitioning can be used to account for radial diffusion in the ISF or physical division of the ISF into two distinct regions (e.g., T-tubules in skeletal muscle). Apportionment of the total ISF volume between these two partitions is controlled by the parameter *e_fVisf2* and the diffusion of tracer between them by the parameter *e_PSisf*. If partitioning of the ISF is not desired, set both *e_fVisf2* and *e_PSisf* to 0.

Typical values of the extracellular tracer parameters, using no axial diffusion and an unpartitioned ISF, are

Name	Value
e_Dp	0.0
e_PSg	1.0
e_Visfp	0.15
e_fVisf2	0.0

Name	Value
e_Disf	0.0
e_PSisf	0.0
e_Czero	0.0

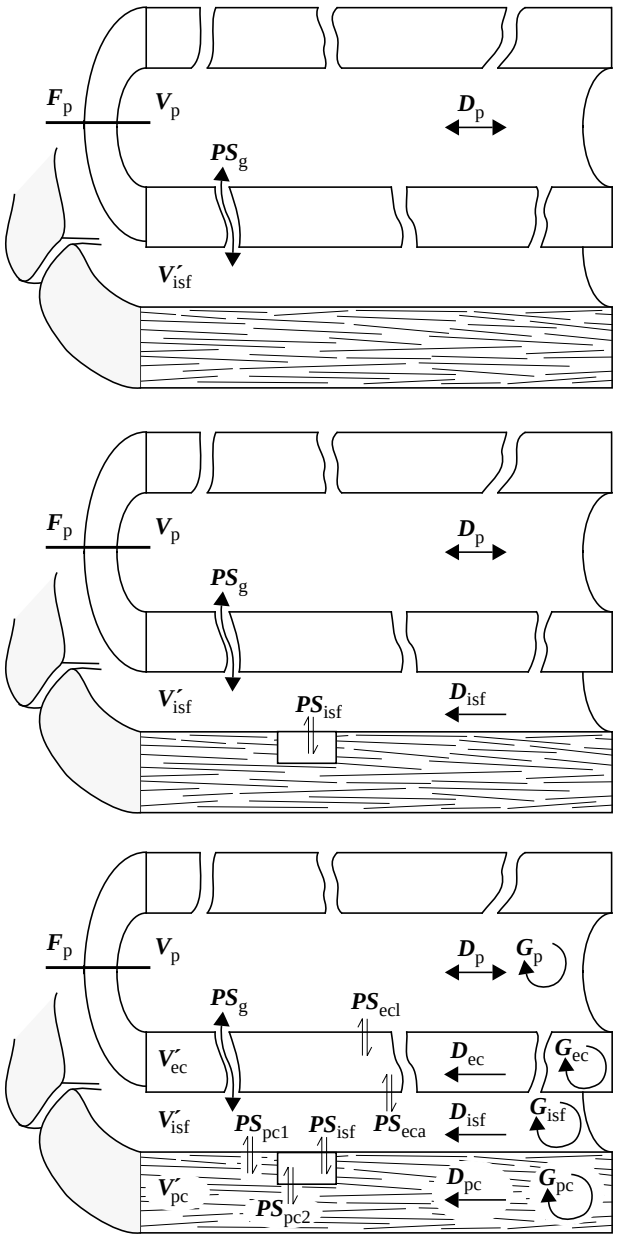


Figure 3.22. BTEX model parameters. *Top:* Vascular tracer. *Middle:* Extracellular tracer. *Bottom:* Permeant tracer.

Permeant tracer

In addition to the plasma and ISF, the permeant tracer enters the endothelial and parenchymal cells (Fig. 3.22, bottom and Fig. 3.23). To not partition the ISF, three parameters must be set to 0: *p_fVisf2*, *p_PSisf*, and *p_PSpc2*.

Typical values of the permeant tracer parameters, using no axial diffusion or consumption and an unpartitioned ISF, are

Name	Value
p_Gp	0.0
p_Dp	0.0
p_PSecl	1.0
p_PSeca	1.0
p_Vecp	0.01
p_Gec	0.0
p_Dec	0.0
p_PSg	1.0
p_Visfp	0.15
p_Gisf	0.0
p_Disf	0.0
p_PSpc1	1.0
p_PSisf	0.0
p_fVisf2	0.0
p_PSpc2	0.0
p_Vpcp	0.60
p_Gpc	0.0
p_Dpc	0.0
p_Czero	0.0

3.4.5. *Vascular and BTEX operator errors and messages*

[TO BE DEVELOPED]

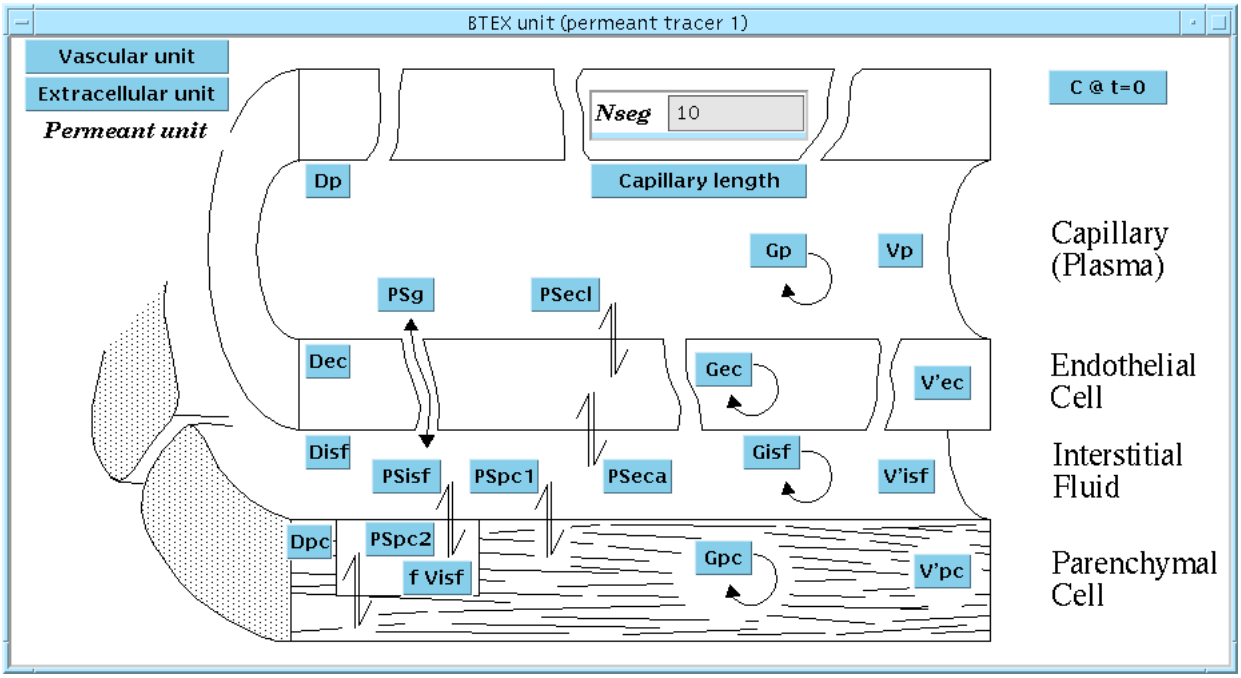


Figure 3.23. Permeant tracer BTEX unit window.

3.5. Displaying Results

3.5.1. *Introduction*

This section deals with the display of results from the MMID4 model. Five types of results are available: heterogeneity (Section 3.5.2), outflow concentrations (Section 3.5.3), tissue contents or residues (Section 3.5.4), residuals (Section 3.5.5), and user defined parameter expressions (Section 3.5.6). The final part of this section deals with errors and messages related to display of results.

3.5.2. *Heterogeneity results*

The parameters that contain the heterogeneity results are shown in the Flow heterogeneity results window.

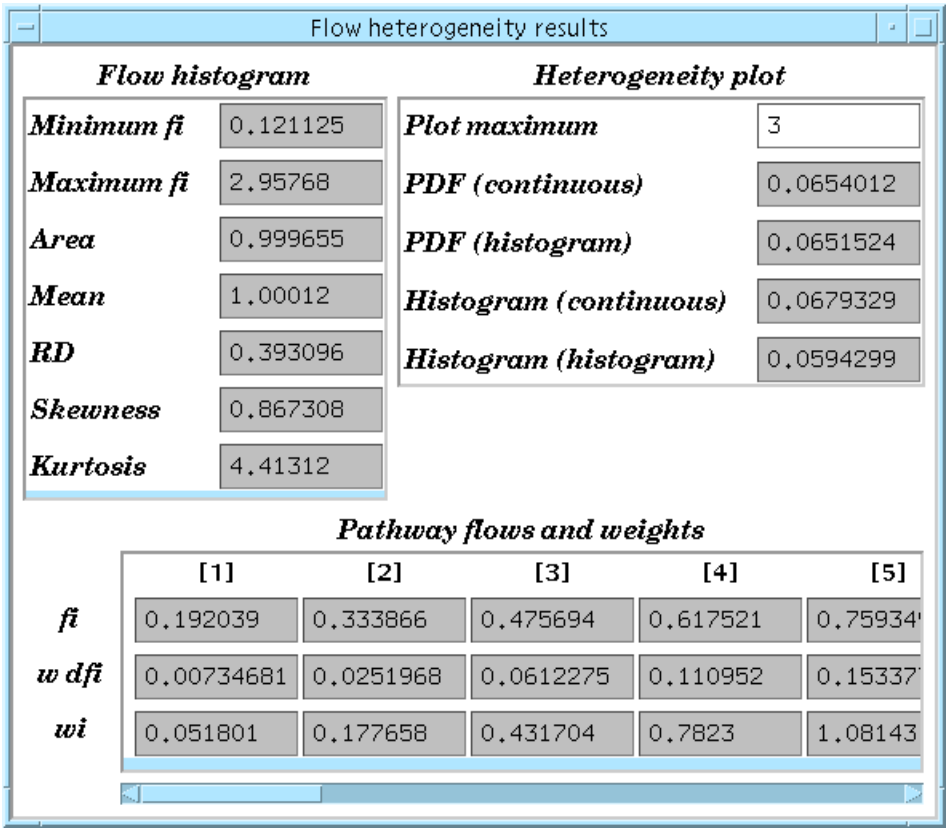


Figure 3.24. Flow heterogeneity results window.

See Section 3.3.2 for the definitions of the parameters shown in Fig. 3.24

The *Flow histogram* parameters and the *Pathway flows and weights* are loaded with single values at the end of the simulation run. The Heterogeneity plot outputs that have a shaded field background are dynamic parameters that are set at each step of the model solution. Scaling and display of these outputs are discussed in Section 3.3.2.

3.5.3. *Tracer concentrations*

Introduction

The parameters that control and contain the tracer concentrations are shown in Table 3.3. Tracer concentrations at the inlet and outlet of the organ and at the outlet of individual operators are available.

Table 3.3: Tracer concentrations parameters

Name	Description	
Cin_v	*Organ inflow concentration:*	*vascular tracer*
Cin_e		*extracellular tracer*
Cin_p		*permeant tracer*
Cout_v	*Organ outflow concentration:*	*vascular tracer*
Cout_e		*extracellular tracer*
Cout_p		*permeant tracer*
Path selection button	Path selector for outflow concentrations: all paths or one selected path	
C_tube_i_v	*Outflow concentration:*	*Inlet tubing (vascular)*
C_tube_i_e		*Inlet tubing (extracellular)*
C_tube_i_p		*Inlet tubing (permeant)*
C_art_v		*Artery (vascular)*
C_art_e		*Artery (extracellular)*
C_art_p		*Artery (permeant)*
C_artl_v		*Arteriole (vascular)*
C_artl_e		*Arteriole (extracellular)*
C_artl_p		*Arteriole (permeant)*
C_cap_v		*Capillary (vascular)*
C_cap_e		*Capillary (extracellular)*
C_cap_p		*Capillary (permeant)*
C_venl_v		*Venule (vascular)*
C_venl_e		*Venule (extracellular)*
C_venl_p		*Venule (permeant)*
C_ven_v		*Vein (vascular)*
C_ven_e		*Vein (extracellular)*
C_ven_p		*Vein (permeant)*
C_tube_o_v		*Outlet tubing (vascular)*
C_tube_o_e		*Outlet tubing (extracellular)*
C_tube_o_p		*Outlet tubing (permeant)*

Organ inflow concentrations

Parameters Cin_v, Cin_e and Cin_p contain the inflow concentrations for the three tracers. These concentrations result from the input function generation discussed in Section 3.2 and are delivered to the upstream end of the inlet tubing.

Organ outflow concentrations

Parameters Cout_v, Cout_e and Cout_p contain the outflow concentrations for the three tracers. These concentrations are measured at the downstream end of the outlet tubing.

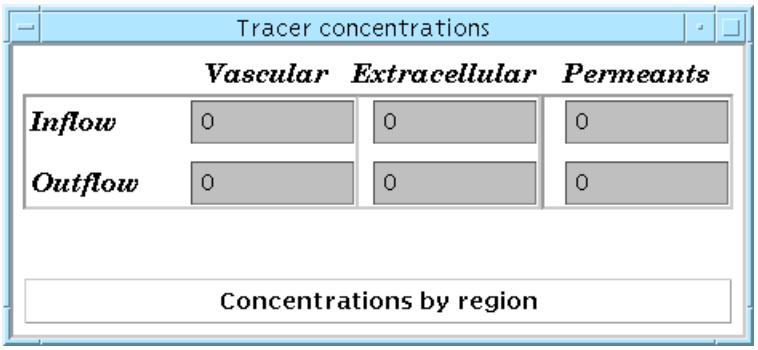


Figure 3.25. Tracer concentrations window.

Outflow concentrations by path

Concentrations of the three tracers at the outlet of individual circulatory components are available in the Concentrations by region window (*Parameters: Model outputs> Concentrations by region*).

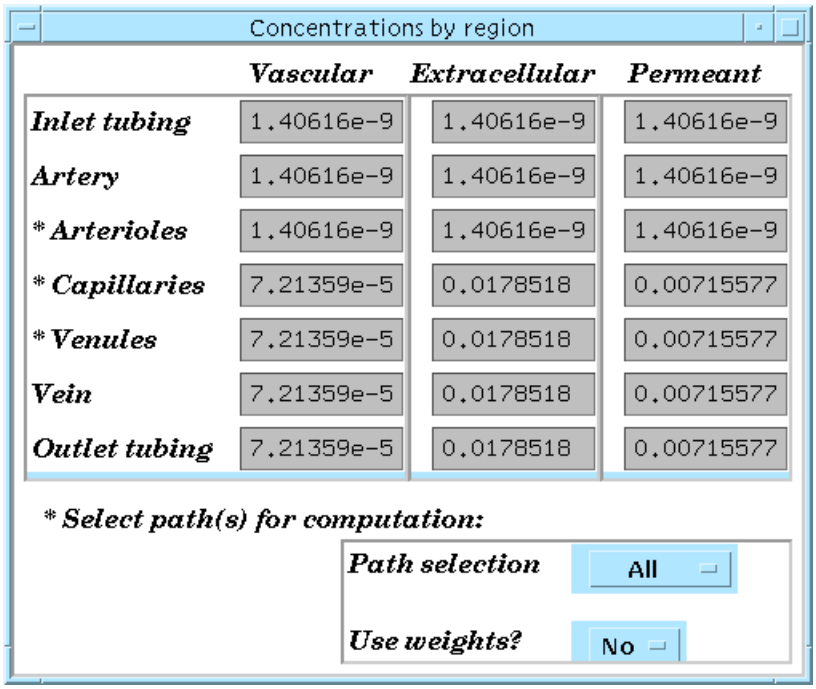


Figure 3.26. Concentrations by region window.

For the arterioles, capillaries, and venules, the concentrations along a single pathway or the average concentration over all pathways are available. This choice is controlled by the *Path selection* button. The path selection status does not, of course, affect the inlet tubing, artery, vein, or outlet tubing. Note that the outlet tubing concentration is the same as the organ outflow for that tracer.

3.5.4. *Tissue contents (residues)*

Introduction

The amount of tracer in the organ, or region of the organ, is denoted by the letter Q (quantity). In MMID4, all residues are calculated by integrating the inflow and outflow curves from the submodels

$$Q(t) = \int_0^t \text{Flow} \cdot (C_{\text{in}}(\tau) - C_{\text{out}}(\tau))d\tau \quad . \tag{3.3}$$

Q for a region that includes several submodels is obtained by adding the individual *Q*'s.

The parameters that control and contain the tracer residues are shown in Table 3.4.

Table 3.4: Tracer contents parameters

Name	Description	
Q_v	*Whole organ residue:*	*vascular tracer*
Q_e		*extracellular tracer*
Q_p		*permeant tracer*
Q_tube_i_v	*Pathway residue:*	*Inlet tubing (vascular)*
Q_tube_i_e		*Inlet tubing (extracellular)*
Q_tube_i_p		*Inlet tubing (permeant)*
Q_art_v		*Artery (vascular)*
Q_art_e		*Artery (extracellular)*
Q_art_p		*Artery (permeant)*
Q_artl_v		*Arteriole (vascular)*
Q_artl_e		*Arteriole (extracellular)*
Q_artl_p		*Arteriole (permeant)*
Q_cap_v		*Capillary (vascular)*
Q_cap_e		*Capillary (extracellular)*
Q_cap_p		*Capillary (permeant)*
Q_venl_v		*Venule (vascular)*
Q_venl_e		*Venule (extracellular)*
Q_venl_p		*Venule (permeant)*
Q_ven_v		*Vein (vascular)*
Q_ven_e		*Vein (extracellular)*
Q_ven_p		*Vein (permeant)*
Q_tube_o_v		*Outlet tubing (vascular)*
Q_tube_o_e		*Outlet tubing (extracellular)*
Q_tube_o_p		*Outlet tubing (permeant)*

Whole organ residues

The whole organ residues for the three tracers are Q_v, Q_e and Q_p, which can be viewed in *Parameters: Model outputs> Tracer contents*. The user can

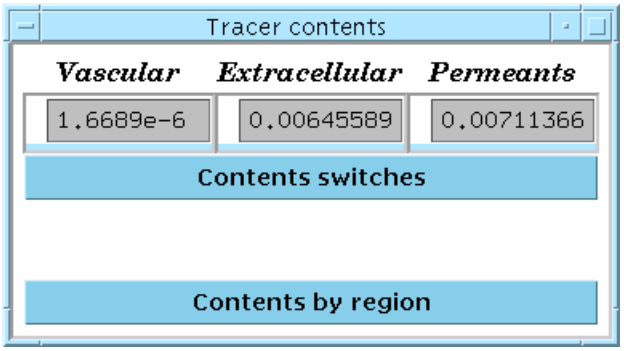


Figure 3.27. Tracer contents window.

specify which vascular components are included in the whole organ residue calculations. Selections are controlled by the contents switches (*Parameters: Model outputs> Contents switches)*.

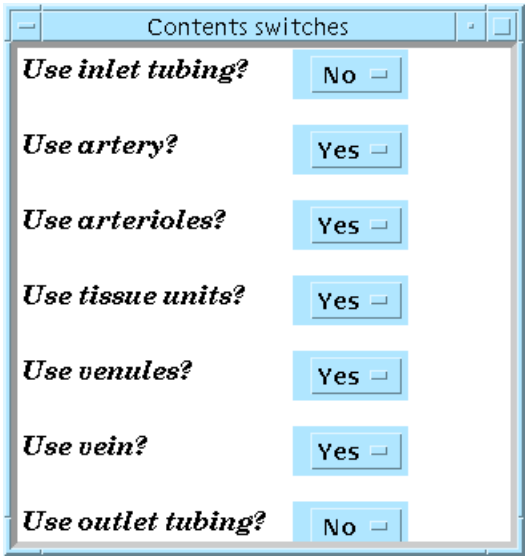


Figure 3.28. Contents switches window.

The example shown in Fig. 3.28 contains only the actual vasculature in the whole organ residue calculation, since the contents of both the inlet and the outlet tubing have been turned off.

Pathway residues

The amounts of the three tracers in individual circulatory components are available in the Contents by region window (*Parameters: Model outputs> Contents by region*).

For the arterioles, capillaries, and venules, the amounts in a single pathway or the total in all pathways are available. This choice is controlled by the path

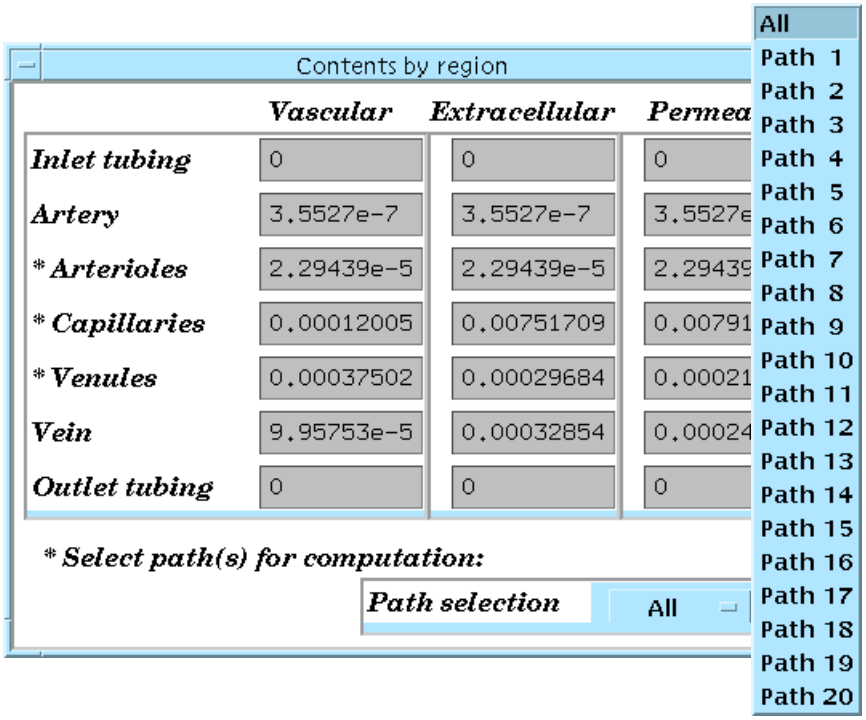


Figure 3.29. Contents by region window with the path selection button activated.

selection button, which does not, of course, affect the values reported for the inlet tubing, artery, vein, or outlet tubing.

3.5.5. *Residuals*

Introduction

In contrast to residue curves, residuals are the difference between a model curve and a data curve:

$$\text{Residual}(t) = C_{\text{model}}(t) - C_{\text{data}}(t) \quad . \tag{3.4}$$

Note that the residuals are not normalized and may be either positive or negative. Values of the residual are only calculated at each point in the data curve.

Residual curve parameters

To create a residual curve graph (Fig. 3.30, top), first make sure that a reference data TAC file has already been loaded into XSIM, then select *Residuals config* from the *Model* pull-down menu. Fill in the desired reference data curve name(s) and the parameter name(s) that you wish to compare (Fig. 3.31), and enter ‘1’ for the point weights and TAC weights if equal weighting is desired (See section 8.2.4 of the Interface Reference Manual for a discussion of weighting options). Click on the Residuals button in the main window and a Residual Plot window should appear.

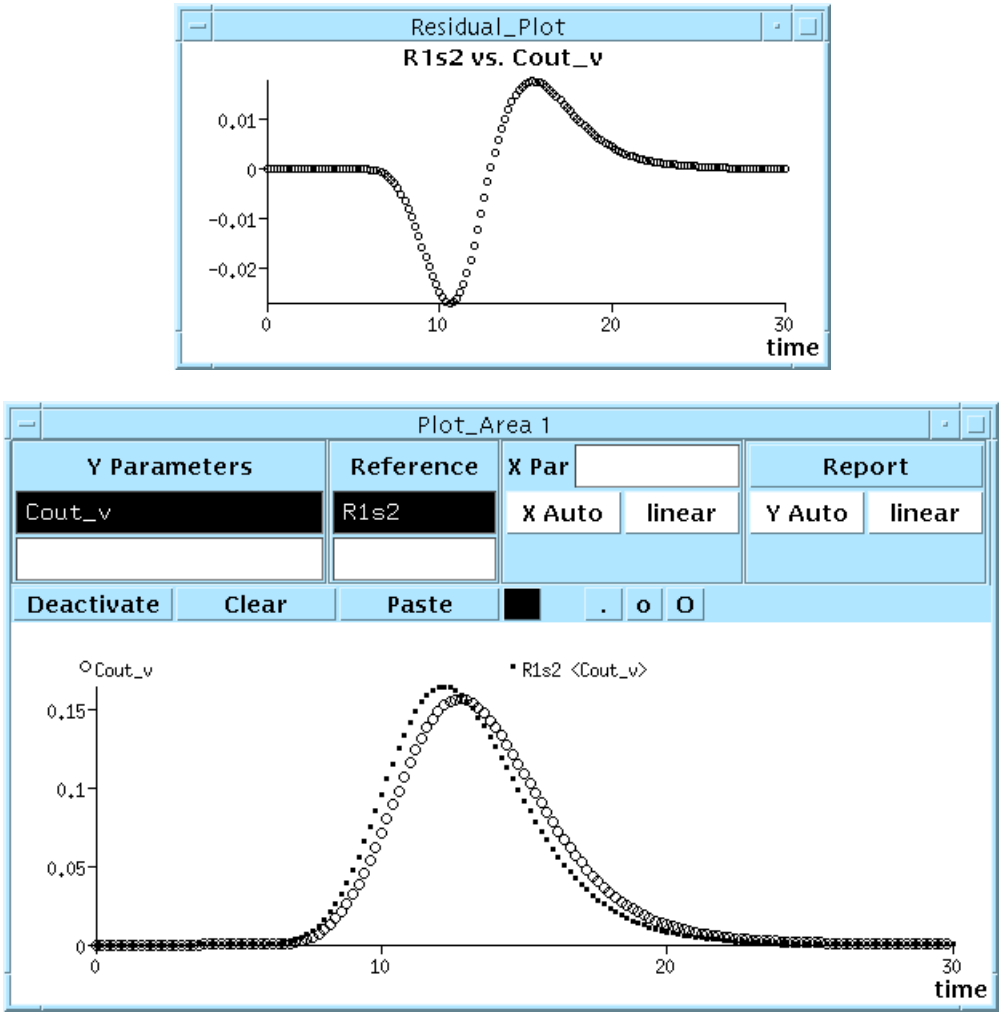


Figure 3.30. Calculation of residual curves. *Bottom*: Data and model curves. *Top*: Residual curve.

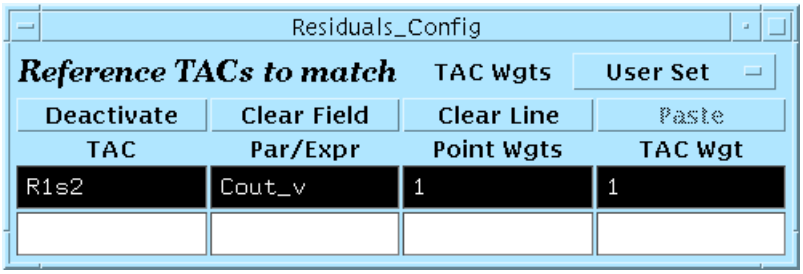


Figure 3.31. Residuals Config window.

3.5.6. *Parameter expressions and scalars*

User defined expressions of parameters may be plotted in either of the plot area windows. For example, the parameter of Cout_v in the previous section may be scaled by 0.5, and offset from zero by 0.1, as in Fig. 3.32. See Appendix B of

the XSIM manual for a listing of the expressions evaluator constants and functions.

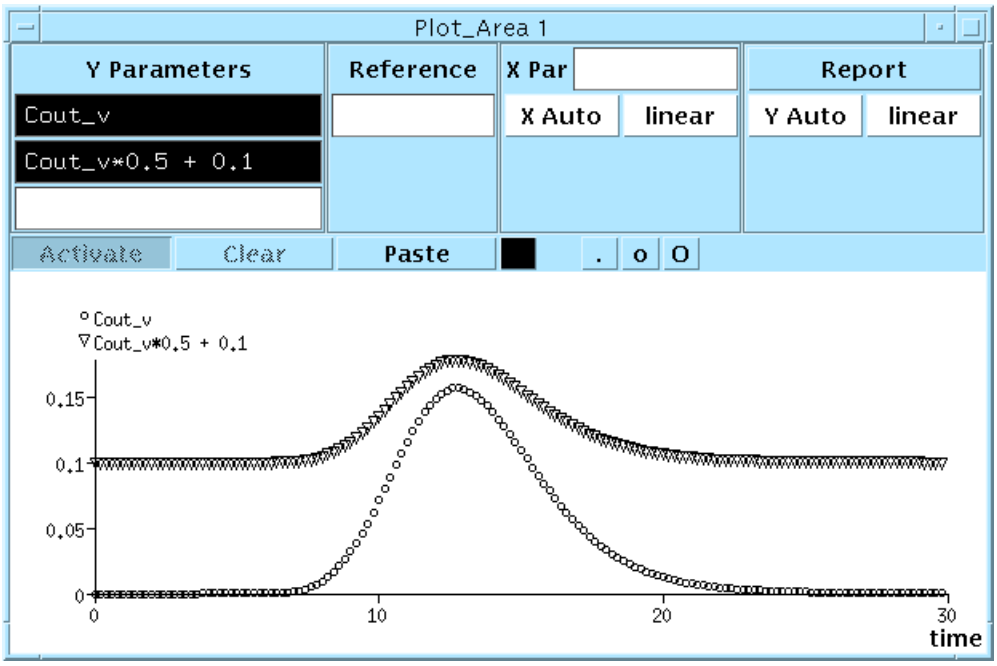


Figure 3.32. Plot of a parameter and the scaled and offset parameter.

If a constant will be used repeatedly and/or often, a scalar may be useful. The set of scalars are simply a set of parameters that are not tied to the model, but can be set like any other parameter, either by entering a value directly or by slaving the scalar to another parameter in the eval field. Scalars are accessed from the Parameters pull-down menu.

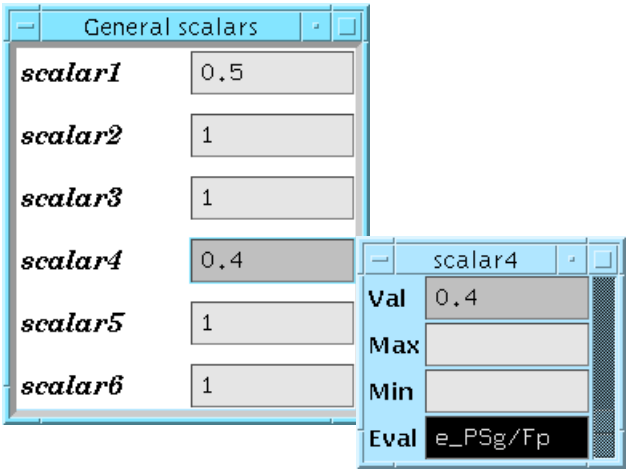


Figure 3.33. General scalars window and the parameter window for scalar4.

In the example of Fig. 3.33, scalar1 has been set to 0.5. One could substitute ‘scalar1’ for 0.5 in the Y parameter field shown in figure 3.8, making the equation *Cout_v*scalar1+0.1*. This scalar could then be optimized on by placing scalar1 in the parameters to vary section of the optimization config window.

Also shown in Fig. 3.33 is an example of using the eval field to make scalar4 equal the ratio of extracellular *PSg* to flow, *Fp*.

3.5.7. *Errors and messages*

[TO BE DEVELOPED]

4. REPORTING BUGS

Bugs in XSIM should be reported by electronic mail to

librarian@nsr.bioeng.washington.edu

Sending an electronic mail bug report may be done from within XSIM. Click on the Help menu on the far right of the main window, and select XSIM bug reports. If you are connected to the World Wide Web, you may also view the XSIM bug report database.

Reports should describe the problem in detail. Where possible, copies of the source code for the user model, a copy of the parameter set used (in ".par" format), and copies of the reference data file should be included in the message. It is also helpful to have copies of any XSIM output files that were produced.

If you do not have electronic mail, send bugs via US mail to

Software Librarian
National Simulation Resource Facility
University of Washington (WD-12)
Seattle, Washington 98195

As a last resort, report bugs via telephone: (206) 685-2005

APPENDIX A. MMID4 TUTORIAL DATABASE CONTENTS

This appendix lists the parameter files in the MMID4 tutorial database for each of the exercises outlined in the tutorial (Section 2). To make your own copy of the tutorial database and required data files, enter the four commands listed below:

```
cd ~
mkdir MMID4tutor
cd MMID4tutor
zcat XSIMHOME/dist/model/MMID4tutor.tar.Z |tar xf -
```

Where *XSIMHOME* is the XSIM installation directory, usually `/usr/local/xsim` (check with your system administrator). Instructions for running MMID4 using the tutorial data base are contained in Section 2.1.4.

Table A.1: Tutorial database contents

Exercise	Notes
Input functions (Parameter file 1input.par)	Using the input function generator
MMID4 configuration (Parameter file 2config.par)	Changing the configuration of MMID4: plasma flow, number of paths, nonexchanging vessels, and blood-tissue exchange units
Plotting results (Parameter file 3plots.par)	Using XSIM to plot different MMID4 output parameters
Mean transit time of the intravascular tracer (Parameter file 4mtt_v.par)	Calculating the mean transit time from flow and volume
Mean transit time of the extracellular tracer (Parameter file 5mtt_e.par)	The effect of PS on mean transit time
Crone calculation of PS (Parameter file 6ps_crone.par)	Calculating the capillary PS with the Crone equation
Influence of PS on exchange (Parameter file 7ps_exch.par)	Examining the effect of flow and capillary PS on the amount of tracer in the ISF
Influence of the parenchymal cell (Parameter file 8p_vs_e.par)	Comparing the distributions of the extracellular and permeant tracers
Influence of the endothelial cell (Parameter file 9endoth.par)	Transport with regions in series and parallel
Choosing an input function (Parameter file 10expinput.par)	Examining the effect of the input function on the shape of the output function and parameter sensitivity
Effect of tracer consumption (Parameter file 11consump.par)	Examining the effect of consumption in different regions
Heterogeneity of flow (Parameter file 12hetero.par)	Using multiple pathways and different models of flow heterogeneity

Table A.1: Tutorial database contents

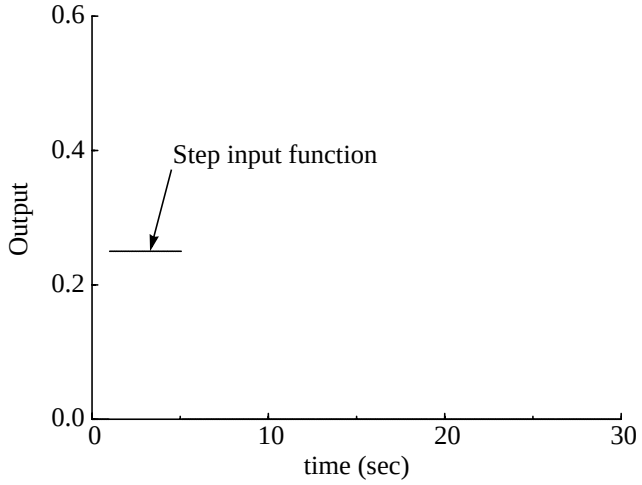
Exercise	Notes
The influence of nonexchanging vessels (Parameter file 13nonex.par)	Adding arterioles and venules to the MMID4 model
Manual parameter optimization (Parameter file 14manopt.par)	Using manual optimization to fit the vascular, extracellular, and permeant tracer curves
Automated parameter optimization (Parameter file 15autoopt.par)	Using automated optimization to fit the three outflow curves
Deconvolution (Parameter file 16decon1.par)	Using deconvolution to generate an input function
Deconvolution with nonexchanging vessels (Parameter file 17decon2.par)	Examining the effect of nonexchanging vessels on deconvolution results
Parameter optimization with noisy data (Parameter file 18noisopt.par)	Examining the effect of noise on parameter optimization
Deconvolution with noisy data (Parameter file 19noisdec.par)	Examining the effect of noise on deconvolution

APPENDIX B. PLOTS FROM SELECTED TUTORIAL EXERCISES

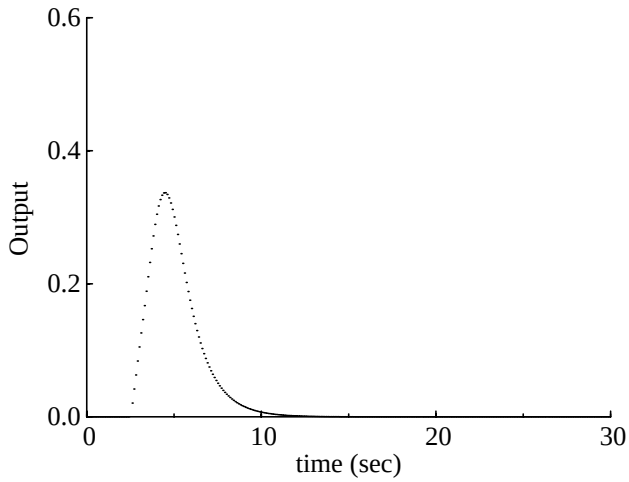
This appendix shows the plots that should be produced by several of the tutorial exercises. The plots shown here will not be identical to those produced online; the model and data curve symbols will differ, and additional labels and annotation have been added. The magnitudes and shapes of the simulation output and data curves should, however, be the same as in the tutorial graphs.

Exercise: Input functions (Parameter file 1input.par)

Default parameters:

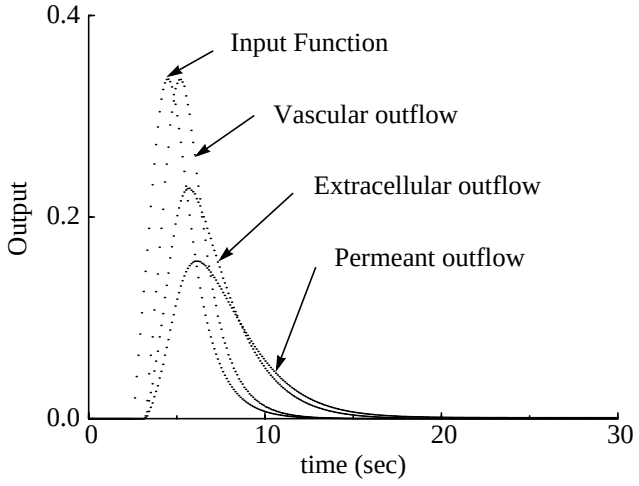


Lagged normal density function:



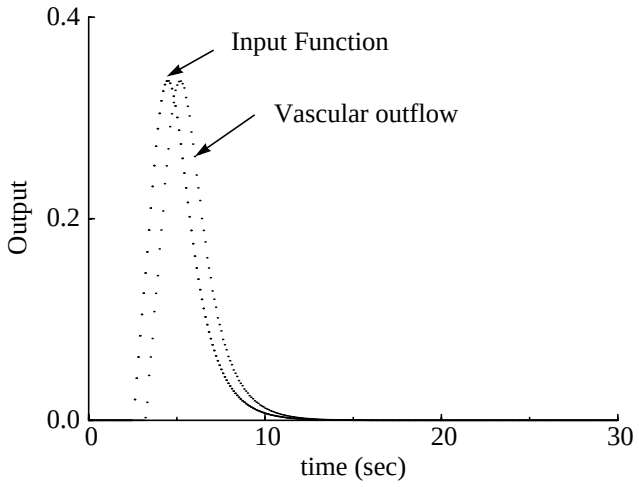
Exercise: MMID4 configuration (Parameter file 2config.par)

Default parameters:

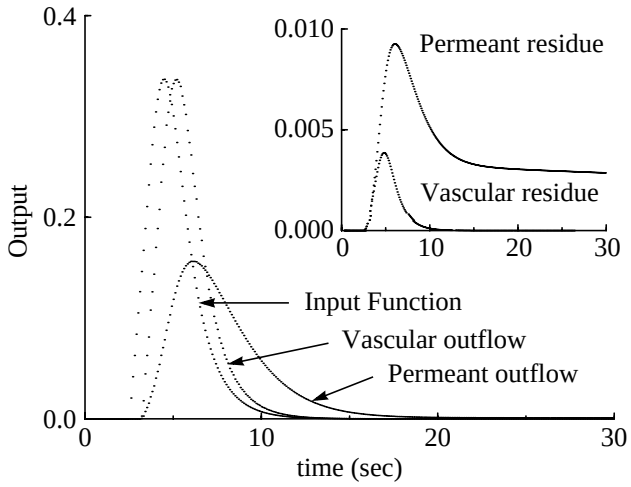


Exercise: Plotting results (Parameter file 3plots.par)

Default parameters:

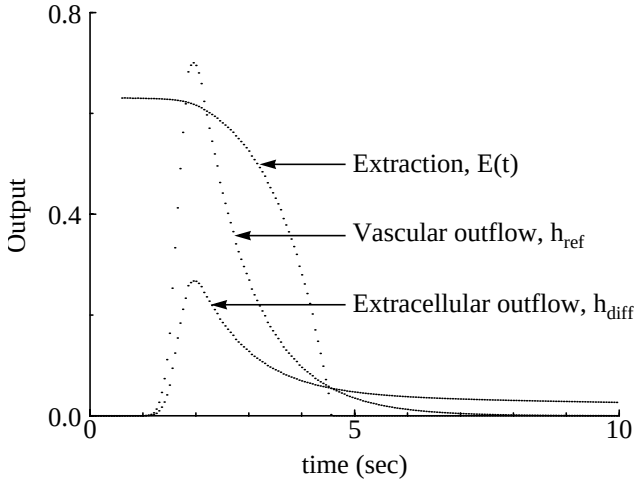


Permeant outflow, vascular residue, and permeant residue added:



Exercise: Crone calculation of PS (Parameter file 6ps_crone.par)

Default parameters:

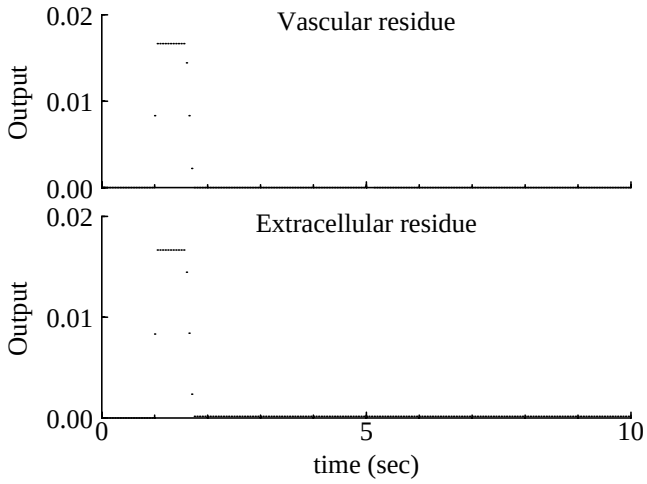


Exercise: Influence of PS on exchange (Parameter file 7ps_exch.par)

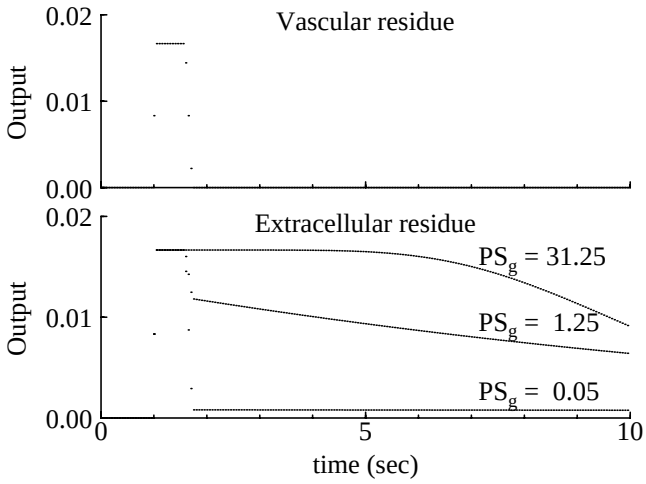
Exercise: Influence of the parenchymal cell (Parameter file 8p_vs_e.par)

Exercise: Influence of the endothelial cell (Parameter file 9endoth.par)

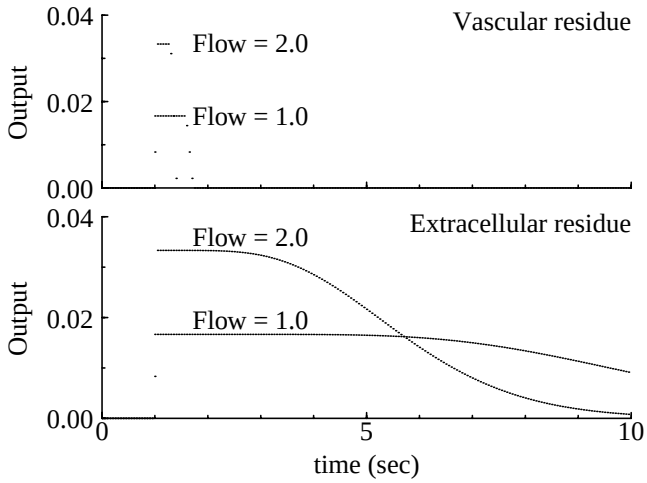
Default parameters:



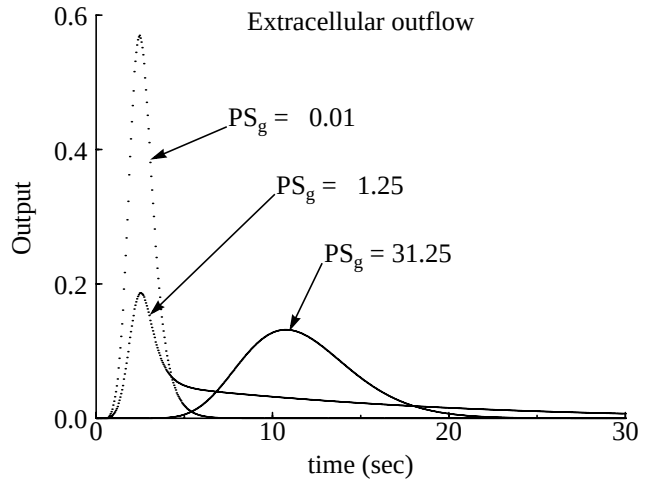
Experiment PS-1:



Experiment PS-2:



Experiment PS-3:



Exercise: Choosing an input function (Parameter file 10expinput.par)

Exercise: Effect of tracer consumption (Parameter file 11consump.par)

Exercise: Heterogeneity of flow (Parameter file 12hetero.par)

Exercise: Manual parameter optimization (Parameter file 14manopt.par)

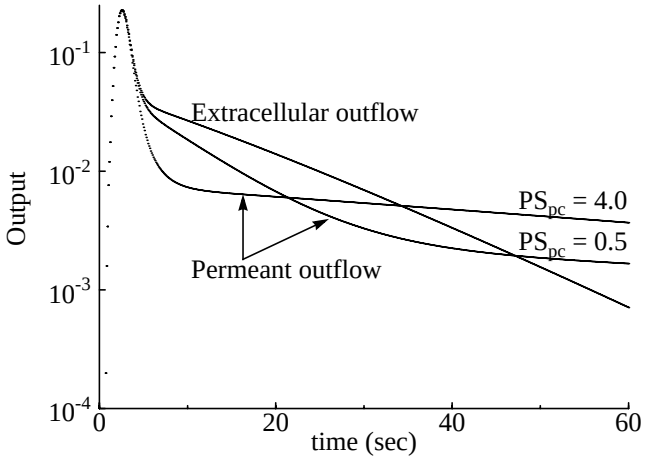
Exercise: Deconvolution (Parameter file 16decon1.par)

Exercise: Deconvolution with nonexchanging vessels (Parameter file 17decon2.par)

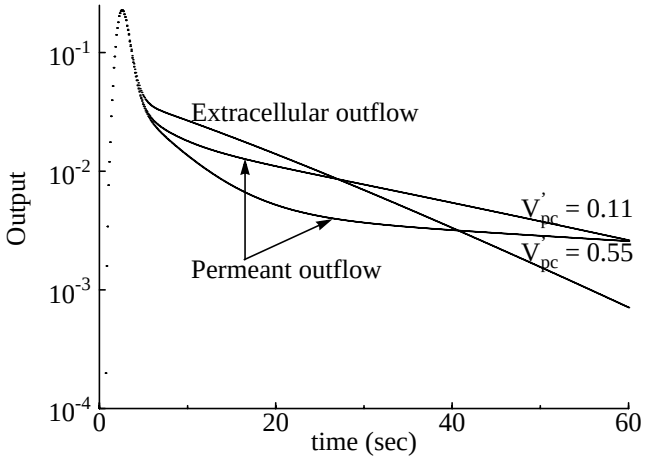
Exercise: Parameter optimization with noisy data (Parameter file 18noisopt.par)

Note: The inset graphs shown below show the fit at the peak on a linear scale. These insets are not present on the tutorial output.

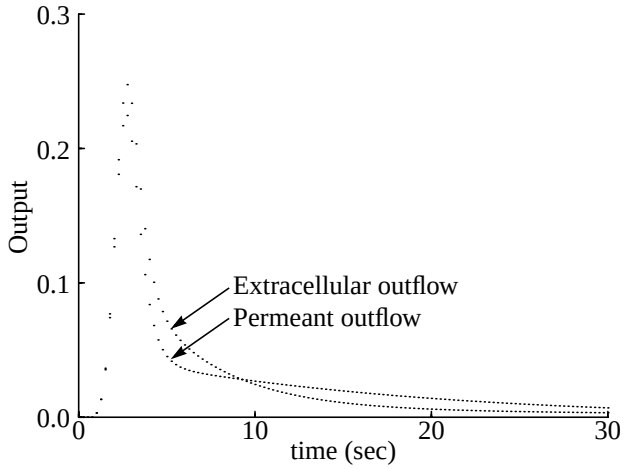
Experiment PC-1:



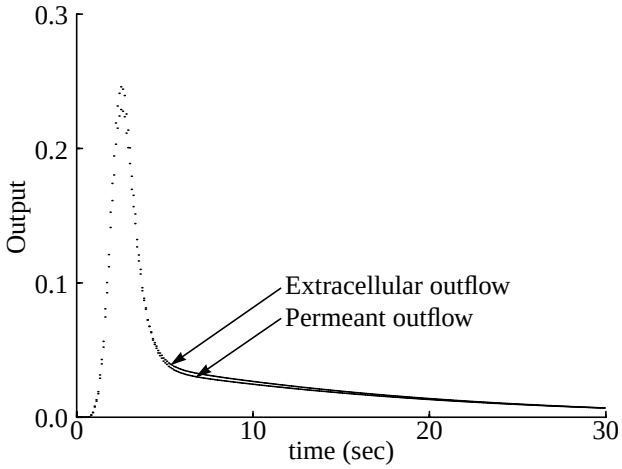
Experiment PS-2:



Experiment EC-1:

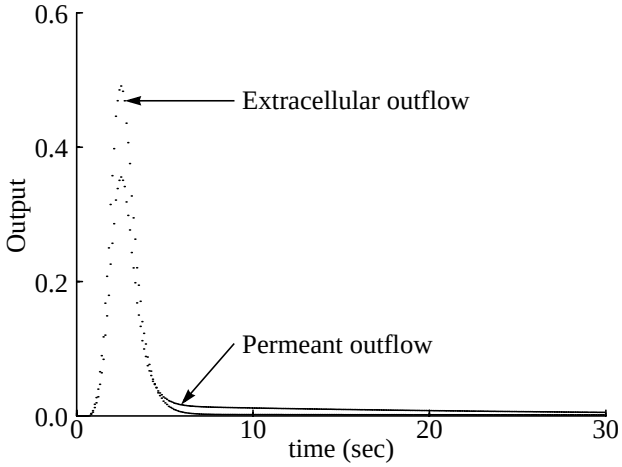


Experiment EC-3:

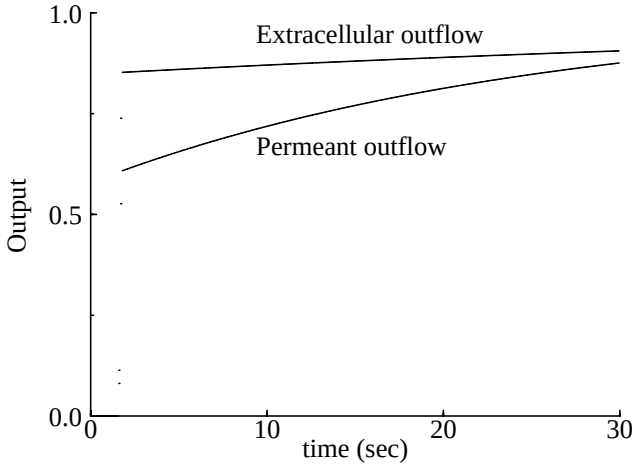


Exercise: Deconvolution with noisy data (Parameter file 19noisdec.par)

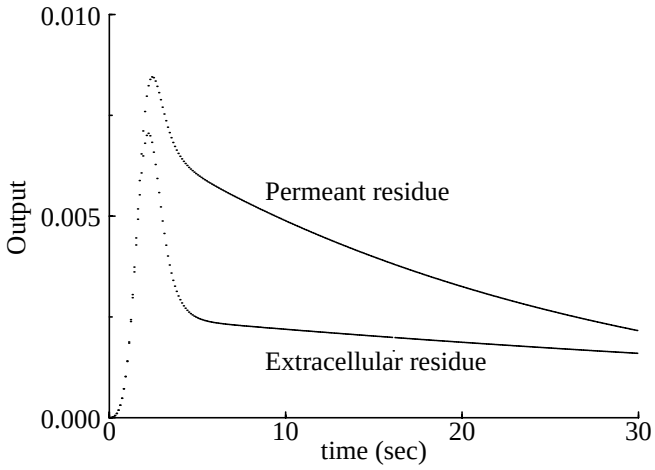
LAGNDC input:



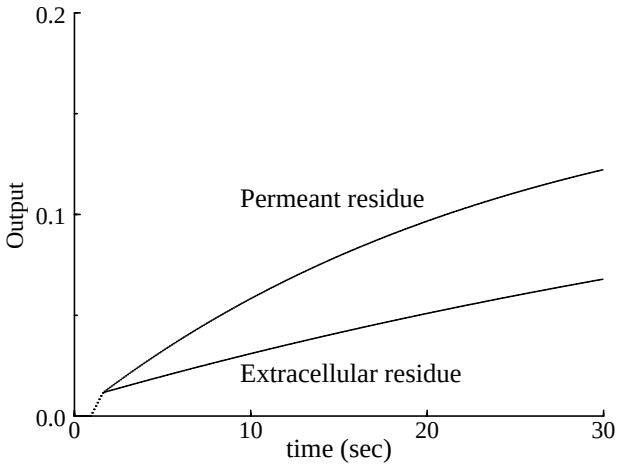
Constant infusion:



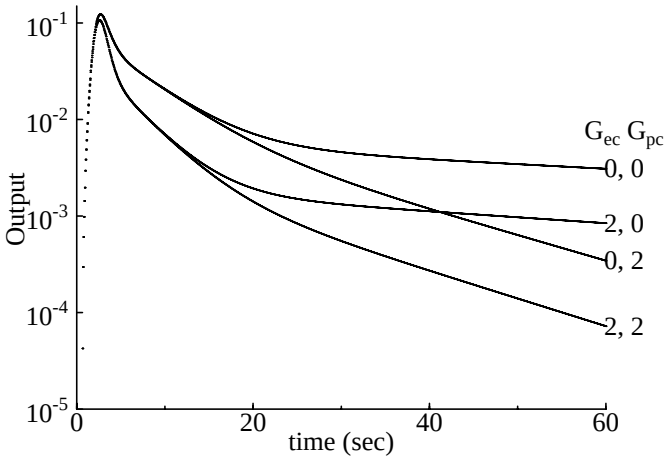
LAGNDC input:



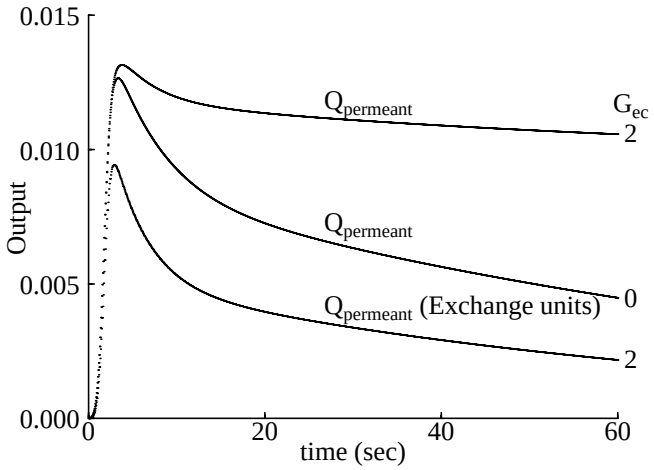
Constant infusion:



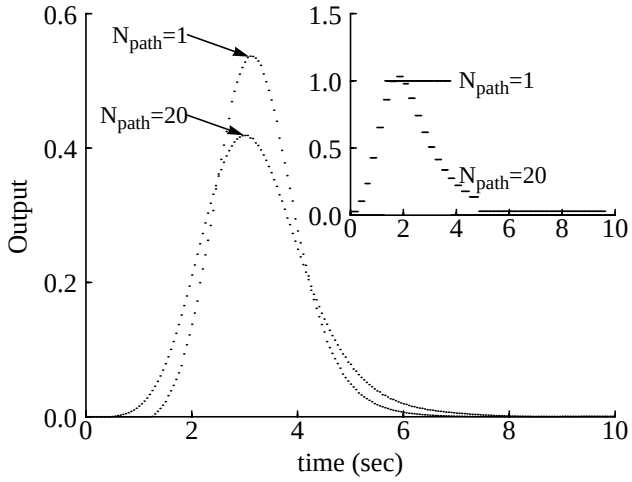
Experiment G-2:



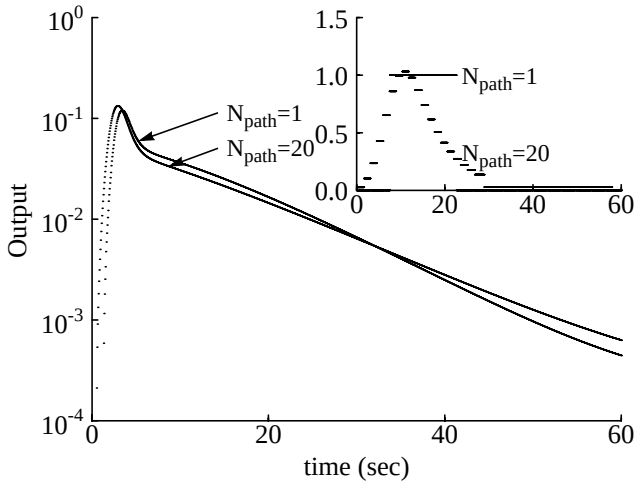
Experiment G-3:



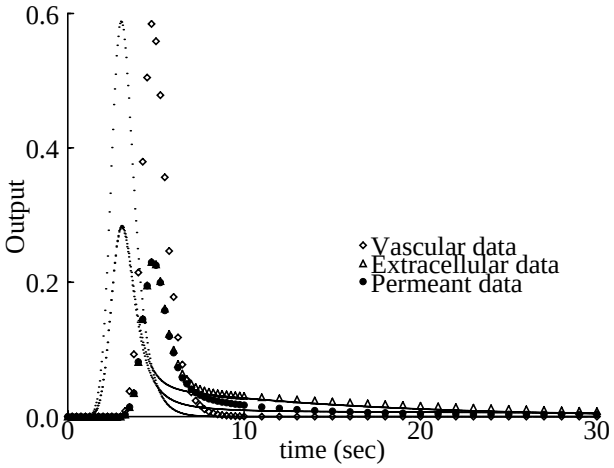
Experiment H-1 (Vascular outflow):



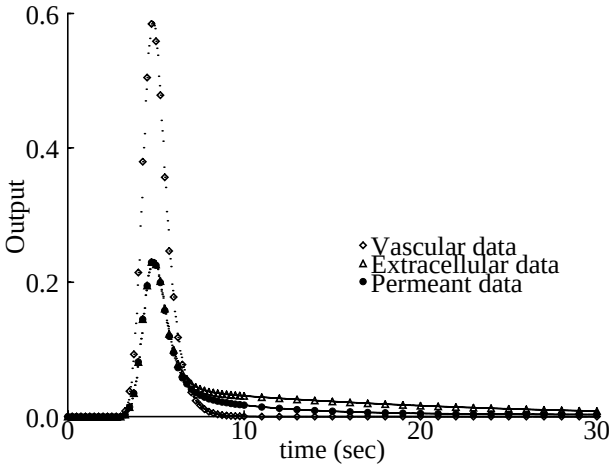
Experiment H-1 (Permeant outflow, PS_{pc} = 0.1):



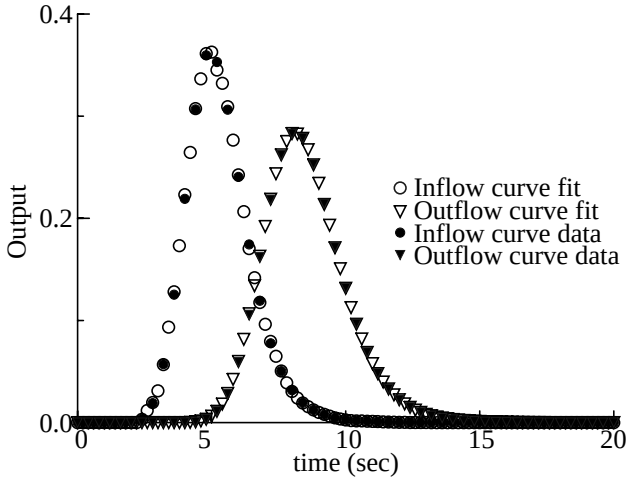
Initial values:



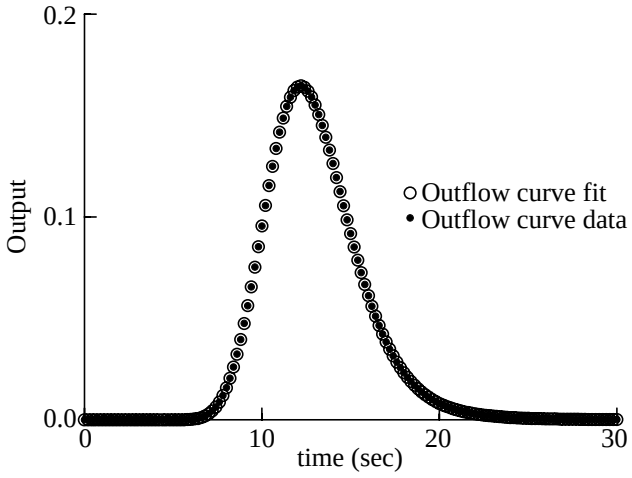
Final fits:



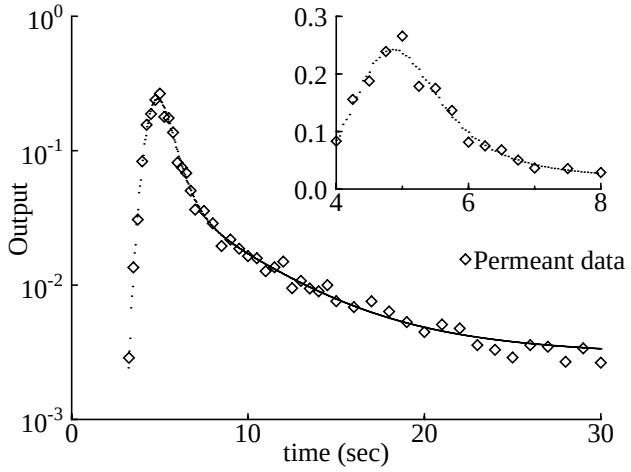
Deconvolution results:



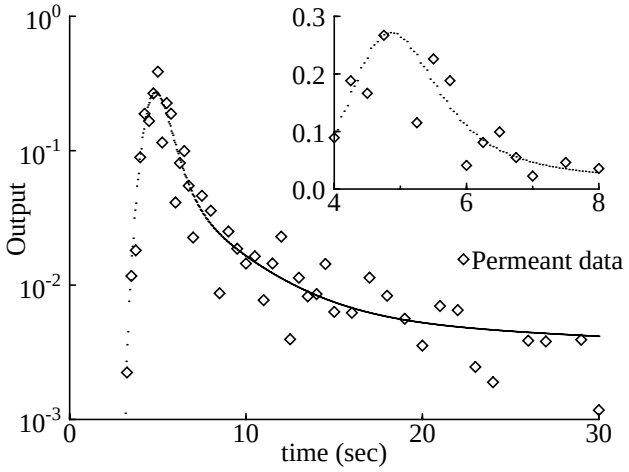
Deconvolution results:



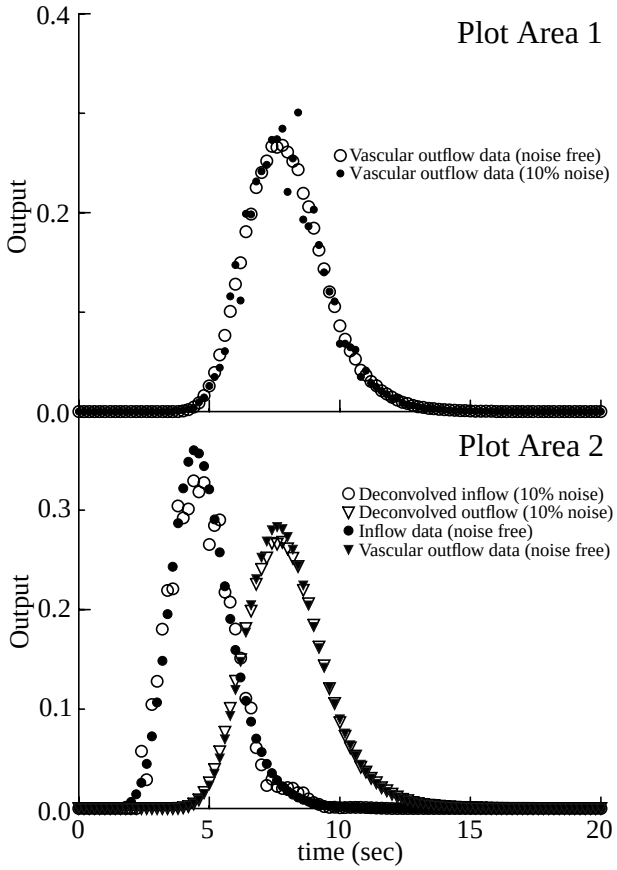
Optimization results (10% noise):



Optimization results (40% noise):



Deconvolution results (10% noise, smoothing=30):



Deconvolution results (40% noise, smoothing=100):